VOLUME XXVIII NUMBER 2

BOTANICAL GAZETTE

AUGUST 1899

THE DEVELOPMENT OF THE MICROSPORANGIA AND MICROSPORES OF HEMEROCALLIS FULVA.[1]

EDWARD L. FULLMER.

(WITH PLATES VII–VIII).

ALTHOUGH *Hemerocallis fulva* has been a frequent subject of study, the results reported in reference to the phenomena of karyokinesis and spore formation do not seem to agree. In the *Cytologische Studien*,[2] this plant is described with multipolar spindles in the reproductive cells during the early division stages, no centrosomes being found in such cells. The division of the reproductive cells of the higher plants studied, according to the results of the several contributors to the *Studien*, is found to differ in both of these respects from the cells of the lower plants as well as from those of animals.

The spore-mother-cells of Hemerocallis are not very favorable for cytological study, the chromosomes being small and the spindles difficult to differentiate on account of a band of dark staining material found close around each nucleus; nor are the spindle threads so thick as those usually found. The dark band may be only an artifact, caused by the chromic acid of the killing solution. The material for this study was killed in chrom-acetic acid, and after being imbedded in paraffin was sectioned 12 and 14 μ thick. The stains used were iron-alum-hæmatoxylin;

[1] Contribution from the botanical laboratory of Ohio State University.

[2] Jahrbücher für wiss. Bot. **30**: 1–268. *pl. 18*. 1897.

Flemming's triple stain; a combination of anilin-safranin with iron-alum-hæmatoxylin; and anilin-safranin followed by picro-nigrosin. The last named gave the best results in staining the incipient spindles.

DEVELOPMENT OF THE MICROSPORANGIA.

A cross section of a very young stamen, at the point where the microsporangia are to be formed, shows merely a rectangular area which consists of epidermal and general tissue cells. Three or four hypodermal cells of each sporangium become differentiated as archesporial cells. These divide by periclinal division, giving rise to the primary sporogenous cells and the primary tapetal layer. The cells of the primary sporogenous tissue multiply rapidly, forming only sporogenous cells. The division of these cells is practically complete when the primary tapetal layer begins to divide. While the sporogenous cells are enlarging the division of the primary tapetal layer takes place, forming a wall layer and an inner layer. The inner layer divides once, forming an intermediate or middle layer, and the layer which develops into the peripheral part of the tapetum, the axial part being derived from the adjacent general tissue.

DIVISION OF THE MICROSPORE MOTHER CELL.

A typical spore mother cell when division begins (*fig. 1*) shows a large nucleus in the center, with radiations in the cytoplasm extending from the nucleus outward toward the cell wall. There is usually a layer of deeper staining cytoplasm next the nucleus, and one or more large nucleoli just within the nuclear membrane. The young spindle is bipolar from its first appearance, each pole being dome-shaped for a time (*figs. 2, 10*). The spindle encloses the nucleus through which the chromosomes are scattered, and the layer of deep staining cytoplasm having moved outward from the nucleus spindle lies wholly within it. The radiations above described appear to be of the same nature and to have the same function as those described in animal cells, such as fish eggs. If they are consumed in spindle formation,

they must be drawn entirely within the dark layer before the nuclear membrane has disappeared.

A careful study was made of preparations containing cells in various stages of division, many of which showed distinctly dome-shaped bipolar spindles just after the nuclear membrane had disappeared. Occasionally a nucleus was observed which seemed to have radiations extending outward, but these were never sufficiently distinct to be considered as forming a multipolar spindle. The spindle was not differentiated by the stains in any case until after the nuclear membrane had almost disappeared, as the dark band obscured the poles. At first these alone would be visible, since the remainder of the spindle could scarcely be distinguished from the nuclear membrane because of their close proximity; the central part of the spindle perhaps being in contact with the nuclear membrane. The spindle fibers at this stage are delicate, the spindle being in process of formation. The fact that the nuclear membrane disappears while the spindle is forming lends support to the theory that the material of the nuclear membrane is consumed in spindle formation.

As the poles separate, the spindle gradually becomes pointed and seems to grow somewhat narrower in the middle portion, so that it is not as wide as was the original nucleus (*figs. 1–7, 10–12*). The chromosomes are drawn into the equatorial plane (*figs. 8, 14*) soon after the spindle becomes elongated to definite points. Small deep-staining bodies which have the appearance of centrosomes are often found at the poles in the various stages of both the first and second divisions (*figs. 3, 7, 9, 13*).

In the second division of the spore-mother-cell the two nuclei may divide successively (*fig. 22*), but almost always simultaneously (*figs. 14, 16*), the spindles usually being parallel to one another (*fig. 15*), but occasionally obliquely placed (*fig. 16*), and often persisting even after division is complete (*fig. 17*). The phenomena of the first and second divisions are the same, except that in the latter the nuclei are smaller and the dark band is not so conspicuous.

Four microspores (*figs. 18, 19*) are usually produced from each spore-mother-cell, but occasionally five, six, and even eight are formed (*figs. 20–27*). Miss Lyon[3] also found this peculiarity in *Euphorbia corollata*, which in the case of Hemerocallis has been known for some time. The extra nuclei were first described as being produced by one or more of the four tetrad nuclei dividing by karyokinesis. Recently, however, they have been described by Juel[4] as being formed from chromosomes which became isolated in one of the divisions of the spore-mother-cell. Many tetrads having supernumerary nuclei were examined, only a few of which seemed to show definitely how the extra number of nuclei might have arisen. In all cases where the origin was indicated by spindles or otherwise they seemed to have been produced by a subsequent karyokinesis. The extra nuclei in *figs. 20* and *21* may be a result either of the indirect division of one of the cells of the tetrad, or of the threefold division of one of the nuclei at the second division. The division of one nucleus into three or more is quite common in animal cells in case of pathological tissue. Wilson,[5] in speaking of cases of pathological mitosis, says:

> The abnormal forms of mitoses are arranged by Hanseman in two general groups, as follows: (1) *asymmetrical mitoses*, in which the chromosomes are unequally distributed to the daughter cells, and (2) *multipolar mitoses*, in which the number of centrosomes is more than two, and more than one spindle is formed. . . . . Lustig and Galeotti ('93) showed that the unequal distribution of chromatin is correlated with and probably caused by a corresponding inequality in the centrosomes, which causes an asymmetrical development of the amphiaster.

The same author refers to the discovery of Galeotti that asymmetrical mitoses may be artificially produced in the epithelial cells of salamanders by treatment with various drugs. Guignard[6] finds very irregular and also multipolar spindles in the

[3] A contribution to the life history of *Euphorbia corollata*. BOT. GAZ. **25**: 418–425. 1898.

[4] Jahrbücher für wiss. Bot. **30**: 205–226. 1897.

[5] The cell in development and inheritance, 67–68. 1896.

[6] Ann. Sci. Nat. Bot. VIII. **6**: 177–220, *pl. 9–11*. 1898.

spore-mother-cells of *Nymphæa alba* and *Limodorum abortivum*, and also in the tapetal cells of *Magnolia Yulan*. A number of spindles from cells of the above-named plants and also from the cells of *Nuphar luteum*, having well defined centrospheres and radiations around the poles, are shown.

If the fifth nucleus in *figs. 20* and *21* was formed by the division of one of the tetrad nuclei, the division was probably normal. In this case the presence of spindles suggesting tripolar mitosis would be explained by the persistence of spindle structures as in *fig. 17*. *Fig. 22* is similar to *figs. 20* and *21*, except that one of the cells resulting from the first division of the spore-mother-cell did not divide. *Fig. 23* may have arisen by any one of the processes described under *figs. 20* and *21*, but since there is no spindle connecting either of the two smaller nuclei with one of the larger, they may have arisen from a chromosome which was isolated in the first division of the spore-mother-cell, forming a nucleus and afterward dividing. In this case but one of the cells resulting from the first division could have divided, as there are only three other nuclei present. However, the three are nearly uniform in size, and all have the general appearance of tetrad nuclei, and not the appearance of a nucleus formed in the first division, as the elongated nucleus in *fig. 22*, which has the usual shape of such nuclei.

Fig. 24 shows four nuclei, two of which are dividing by karyokinesis. The appearance and position of the cell walls, as well as the unequal size of the nuclei, suggests that the two nuclei resulting from the first division of the spore-mother-cell were of unequal size. By the subsequent division of the nuclei thus formed two pairs of nuclei resulted; the larger pair of which are again dividing by karyokinesis. This figure may also be explained by supposing the two smaller nuclei to have been produced from an isolated chromosome. In this case the spore-mother-cell could have divided but once, and is now dividing a second time. One of the five nuclei in *fig. 25* is dividing. The source of this nucleus cannot be determined with certainty; but it may have been formed either by the indirect division or by the fragmentation

of one of the tetrad nuclei, more probably the smallest one. The fact that several, perhaps the normal number of chromosomes are present opposes the view that it arose from an isolated chromosome. Six nuclei are found in *fig. 26*, the origin of four being shown by spindles. The two extra nuclei are quite small, containing but little chromatin, and may have been produced by isolated chromosomes as above described. However, if they were produced in this manner the smallest of the four nuclei must have arisen by pathological mitosis by an unequal distribution of chromatin, for it is very small and contains but little chromatin. *Figs. 27* and *28* show tetrads with supernumerary cells. The cells of such tetrads are seldom of equal size, one or more of the cells usually containing a small nucleus.

THE DEVELOPMENT OF THE MALE GAMETOPHYTE.

The microspores soon after their separation and rounding off acquire the general shape and characteristic markings of the mature pollen grain, gradually becoming very much enlarged (*figs. 28, 30*). A single elongated nucleus, which seems to have a small amount of chromatin, is found in the center of the spore until growth is far advanced; after which the division into generative and tube nuclei occurs (*fig. 30*). Until recently it was thought that the tube nucleus never divides, but Chamberlain[7] found many cases of such division in *Lilium tigrinum* and *L. auratum*. Smith[8] found the same thing to be of frequent occurrence in *Eichhornia crassipes*. Hemerocallis also shows this peculiarity (*figs. 31–35*), many pollen grains having two, and a few having three, four, or even six tube nuclei. These were formed by direct division in all the cases that I observed, as were those reported by Chamberlain. *Fig. 33* shows a very irregular tube nucleus.

If the tube nucleus is the homologue of the cover cell of the antheridium of Marsilia, and represents the wall of an antheridium, it might sometimes divide through reversion to its more

[7] BOT. GAZ. **23**: 423–430. 1897.

[8] BOT. GAZ. **25**: 324–337. 1898.

primitive condition. Since extra tube nuclei are produced only by fragmentation, so far as known, such division may represent a pathological condition. It is possible that direct division in the higher plants never represents anything more.

SUMMARY.

1. Three or four hypodermal cells of each sporangium become differentiated as the archesporial cells. The wall of a sporangium consists of three layers exclusive of the epidermis. The tapetum is a physiological rather than a morphological structure, the peripheral part being organized from the wall layers and the axial part from the general tissue.

2. The spindle appears bipolar from its first appearance, being dome-shaped in the early stages. No trace of multipolar spindles was observed. The spindles often persist for a considerable time after division is complete. Bodies having the appearance of centrosomes are frequently seen at the poles.

3. The origin of the supernumerary microspores was not absolutely determined. In many cases where their origin was indicated by spindles or otherwise they appeared to arise by the indirect division of one of the tetrad nuclei.

4. The tube nucleus frequently divides by direct division, forming sometimes as many as six or eight nuclei.

In conclusion I wish to express my thanks to Dr. W. A. Kellerman and Mr. J. H. Schaffner for valuable assistance and criticism.

COLUMBUS, O.

EXPLANATION OF PLATES VII AND VIII.

The figures were drawn with a camera lucida and are reduced to three eighths their original size. *Fig. 32* was drawn with a Bausch and Lomb $\frac{1}{12}$ objective and a Leitz no. 2 ocular (× 1200); *figs. 3, 4, 9, 10, 13, 15, 21, 22, 23* with a Leitz $\frac{1}{16}$ objective and a Leitz no. 4 ocular (× 1600); all others with a Bausch and Lomb $\frac{1}{12}$ objective and a Zeiss no. 6 ocular (× 1600).

PLATE VII.

FIG. 1. Microspore mother-cell in very early stage of first division, showing dark band around nucleus and cytoplasmic radiations extending outward to the cell wall.

FIG. 2. Loose mother skein stage, showing a dome-shaped bipolar spindle which is situated entirely within the dark band.

FIGS. 3–5. Successive division stages, showing centrosome-like bodies at poles of dome-shaped spindles.

FIG. 6. Same as above, but having no bodies visible at the poles.

FIG. 7. Mother star stage; spindle pointed, having centrosome-like bodies, and with radiations at lower pole.

FIG. 8. Metakinesis stage; microspores separated.

FIG. 9. Near close of metakinesis; centrosome-like bodies and radiations present.

FIG. 10. Early stage of second division, showing dome-shaped spindle with centrosome-like bodies; dark band not so prominent as in first division.

FIG. 11. Showing one cell in a later stage of division than is the other.

FIGS. 12–14. Successive division stages.

FIG. 15. Loose daughter skein stage, showing an isolated chromosome; spindles parallel.

FIG. 16. Same as above, showing spindles lying obliquely to one another.

FIG. 17. Tetrad having nuclei variable in size and showing persisting spindles.

FIGS. 18–19. Later tetrad stages.

PLATE VIII.

FIGS. 20–21. Mother cells with five nuclei, three of which are connected by spindles, showing their common origin.

FIG. 22. Same as above, except that one nucleus did not divide after first division of spore-mother-cell.

FIG. 23. Five nuclei in mother-cell, two pairs of which are connected by spindles.

FIG. 24. Four nuclei in tetrad, two of which are dividing by karyokinesis.

FIG. 25. Mother-cell with five nuclei; one is dividing by karyokinesis.

FIG. 26. Mother-cell with six nuclei; the origin of two not shown.

FIGS. 27–28. Five- and six-celled mother cells.

FIG. 29. Microspore soon after separation.

FIG. 30. Normal mature pollen grain having one generative and one tube nucleus.

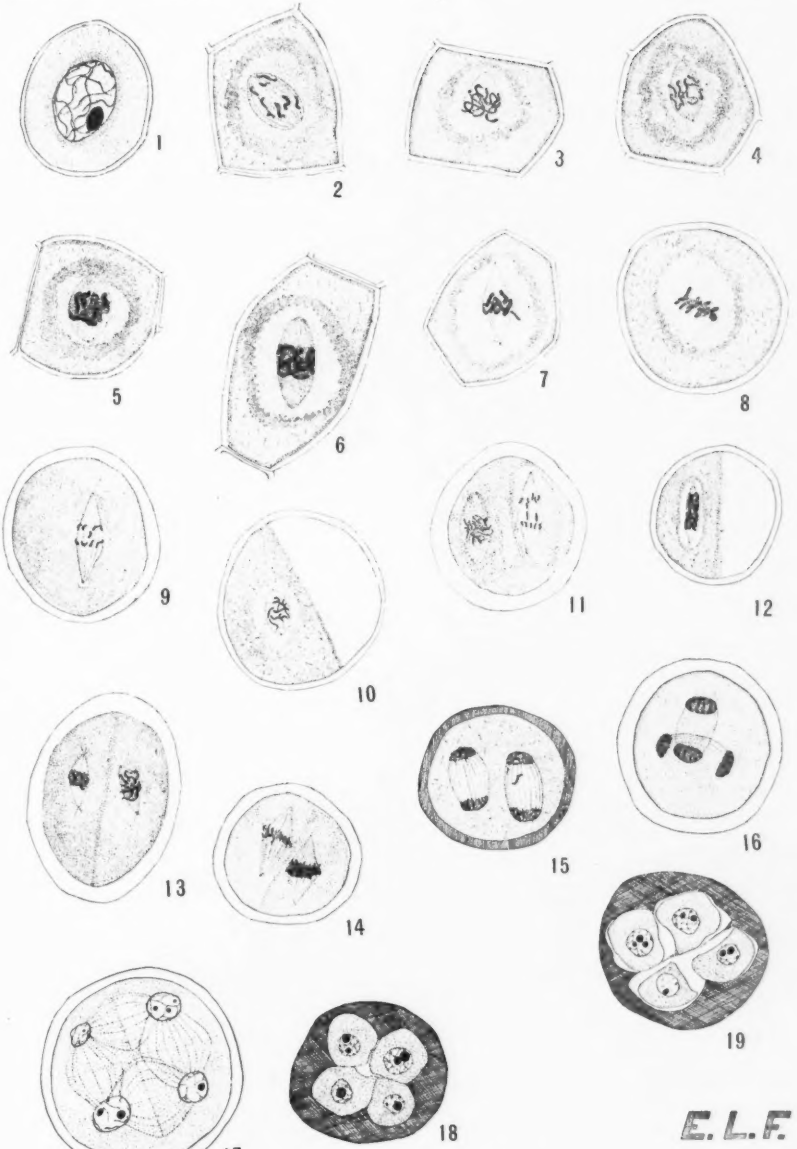
FIG. 31. Tube nucleus of pollen grain dividing by direct division.

FIGS. 32, 34. Pollen grains with one generative and two tube nuclei.

FIG. 33. Pollen grain with a very irregular tube nucleus.

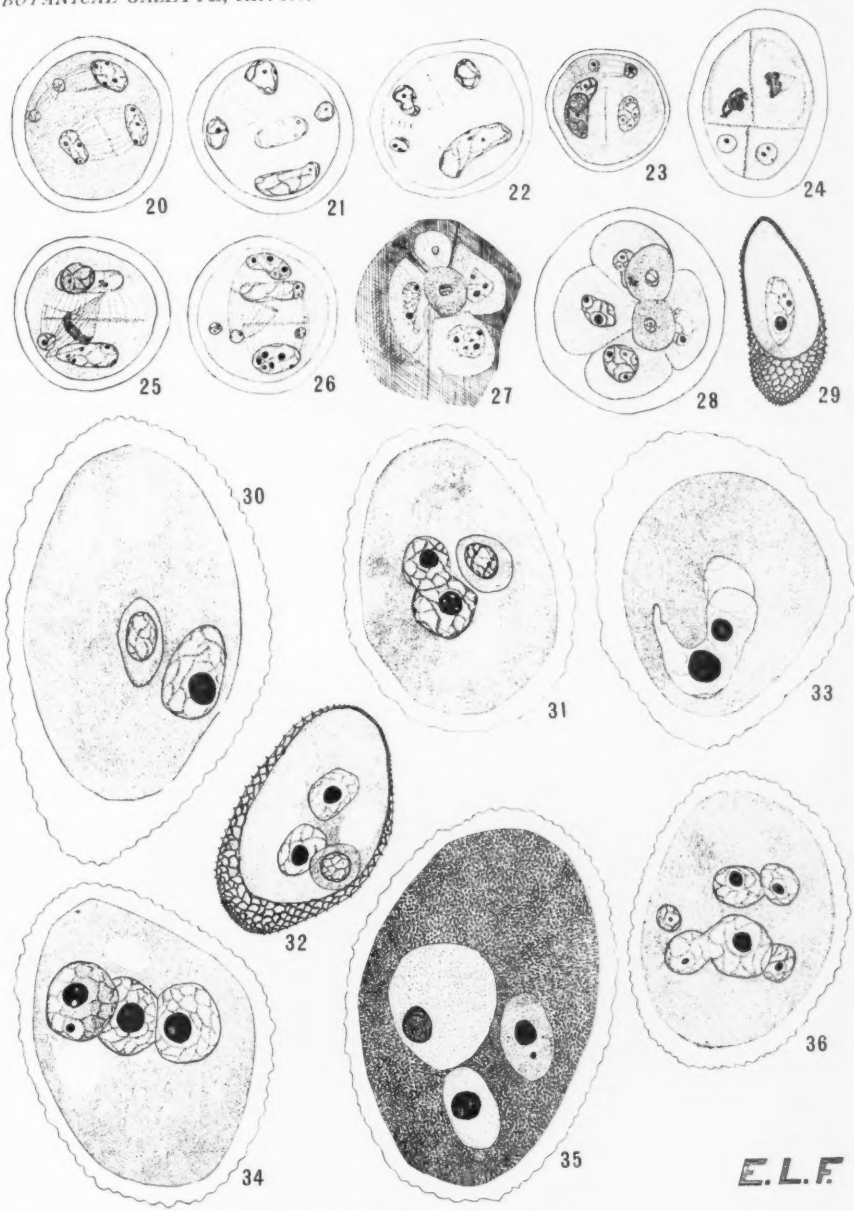
FIG. 35. Pollen grain with three tube nuclei; generative nucleus not shown.

FIG. 36. Six tube nuclei in pollen grain.



E.L.F.

FULLMER on HEMEROCALLIS



FULLMER on HEMEROCALLIS

THE SPORE-MOTHER-CELL OF ANTHOCEROS.

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY. XV.

BRADLEY MOORE DAVIS.

(WITH PLATES IX AND X)

ANYONE who has ever examined sections of the sporogonium of Anthoceros must have been impressed with the extreme beauty of the spore-mother-cells as they are exhibited in all stages of development throughout the length of this structure. The exceptional chloroplast and the various conditions illustrating the division of the cell-contents to form the spores are perhaps the most striking features presented. These led the writer to attempt a detailed examination in the hope that light might be thrown on certain problems that interest the student of plant cytology

The work of Farmer ('94 and '95) is the only contribution of a detailed nature upon the cytology of the Hepaticæ. He did not study Anthoceros but confined himself chiefly to certain thalloid Jungermanniaceæ and reported some very interesting and remarkable conditions. The present investigation does not agree with his accounts of the processes of nuclear division presented by such forms as Pallavacinia, Aneura, Fossombronia, and Pellia. However, at the outset the writer wishes to express no surprise at important differences between such divergent forms as Anthoceros and members of the Jungermanniaceæ. The researches of the past few years in cytology have certainly taught us to be wary of generalizing upon the conditions illustrated by particular cases.

This investigation deals with the single species *Anthoceros laevis* L., the material being collected at Woods Hole, Mass., where were conducted the preliminary studies and experiments on methods of fixation. The best results were obtained

from specimens that had been fixed for twenty-four hours in a weak Flemming solution of the following formula: 1 per cent. chromic acid 25^{cc}, 1 per cent. glacial acetic acid 10^{cc}, 1 per cent. osmic acid 10^{cc}, water 55^{cc}. Much material, however, was killed in other ways, sectioned and stained to determine as precisely as possible the merits of Flemming and the disadvantages of other killing agents. The results of these experiments are brought together at the end of the paper and present some interesting data.

Sections were cut 5 and 7.5 μ thick from material embedded in paraffin and for the most part stained after Flemming's method with safranin, gentian violet and orange G. Excellent results were obtained with iron alum and haematoxylin employed according to the principles of Haidenhain. Acid fuchsin followed by methyl green or gentian violet presented interesting examples of differential staining, but the results were not as good as those given by Flemming's triple stain.

At the outset it will be well to state that the number of chromosomes for *A. laevis* appears to be four and eight in the gametophyte and sporophyte respectively. The count in the gametophyte is based solely upon observations of many mitotic figures in the cells of a developing antheridium, but it agreed with the number presented in the nuclear figures in the spore-mother-cell. The number eight for the sporophytic generation was presented quite frequently and in different regions of that structure. Thus the archesporium just before the differentiation of the spore-mother-cells, the tissue below the archesporium, and the cells of the columella afforded many nuclear figures in which a count could be made with reasonable accuracy.

The attractiveness of the sporogonium of Anthoceros as a subject for study lies largely in the fact that in one structure there may be present a very complete series of stages from the time when the spore-mother-cell is differentiated until the spores are fully ripe. It is possible from this fact to relate to each other with great exactness the events that take place as the spore-mother-cell matures.

As is well known, the sporogonium of Anthoceros grows from a region of embryonic or meristematic tissue in the slightly constricted portion of the structure just above the foot. From this region there rises a central columella around which are arranged in the form of a hollow cylinder the cells destined to develop into spores and sterile cells (elaters). Recently published accounts of the structure and development of the sporogonium by Campbell (**'95,** pp. 130–131) describe this hollow cylinder as two layers of cells thick and composed of groups of spore-mother-cells separated by clusters of sterile cells that later become the elaters. The writer's preparations indicate far less regularity of structure than this description would imply. In many cases the spore-mother-cells formed a single layer around the columella and the amount of sterile tissue was very insignificant. Only occasionally was the layer of cells distinctly double and then very irregularly so. There appears, therefore, to be considerable variation in the complexity of the sporogenous tissue in Anthoceros, and the simplest conditions approach very closely the arrangement of cells reported by Campbell (**'98**) for Dendroceros.

The cubical cells of the archesporium frequently contain nuclear figures, but the spindles are very small and unsatisfactory for study. Such a spindle is shown in *fig. 1*, with the chromosomes separating into two groups and about to pass to the poles. The cytoplasm is granular and contains minute vacuoles. The writer was not able to find a trace of the chloroplast in these cells, but looks forward to a further study of this point in living material and with special methods of staining.

The spore-mother-cells become very quickly differentiated by a rapid increase in size. Their nuclei are not at first to be distinguished from those in the cells of the archesporium, for they are small and contain the one prominent nucleolus and an ill-defined network of linin threads. The most conspicuous change in the cell is the appearance in fixed material of the chloroplast. This structure is at first a slightly denser mass of protoplasm than the other contents of the cell, but so small that it

can scarcely be distinguished from differentiated granular regions of the cytoplasm. Indeed, the first clear proof of its presence in my preparations has been the sharp staining of the starch grains (purple with gentian violet) that are found almost immediately in the interior. The reader may see in *fig. 3* such an early stage in the development of the chloroplast, and should notice that the three small starch grains lie in a homogenous matrix of much the same consistency as the surrounding cytoplasm. The chloroplast in cells younger than that shown in *fig. 3* could not be recognized with certainty by the methods of staining employed in the present study.

The chloroplast once differentiated soon becomes the most conspicuous object in the spore-mother-cell by virtue of its size and brilliant differentiation with stains (*figs. 4, 5*). The peculiarities become more apparent as it enlarges. The outline is sharply marked but it has been impossible to establish a distinct bounding membrane. The interior contains beautifully defined starch grains that become very numerous as the chloroplast increases in size and finally fill by far the greater part of the enclosed region. Each starch grain at first occupies a little cavity surrounded by delicate films of protoplasm that appear as strands in the sections figured (*figs. 5–9*). The chloroplast has therefore a honeycomb-like structure, each cavity being entirely or almost entirely filled by a grain of starch.

The structure of the protoplasmic strands and films around the starch grains proved a very difficult subject for study. They are granular and extend on all sides to the boundary of the chloroplast where they merge into the densely staining film at the exterior. But the bounding film grades off insensibly into the cytoplasm and it was not possible to distinguish by any stain reaction the strands inside the chloroplast from the protoplasm outside.

The first division of the chloroplast takes place very quickly after the structure reaches a certain size. Preparation for the process is usually indicated by the chloroplast elongating and bending around the nucleus in the form of a thick crescent.

The phenomenon of division is one of simple constriction in the middle region and final separation of the halves as the furrow deepens around the chloroplast. It is very plain that the line of fission is determined by the furrow and not from the interior of the chloroplast. Therefore if any particular region of the protoplasm is actively concerned in this process one would naturally first seek the agent in the peculiar film bounding the chloroplast. However there is little in the structure of this region to indicate such important activities.

After the first division of the chloroplast the two portions usually separate and pass to opposite ends of the elongated spore-mother-cell (*fig.* 6). Each has a structure quite similar to the parent chloroplast except that the starch grains become even more numerous and prominent and the fine strands and films of protoplasm separating the starch grains are less clearly defined. Soon after this first division of the chloroplast there takes place that peculiar condition of the nucleus termed synapsis, but this phenomenon had best be considered in a later portion of the paper that deals with nuclear activities. There is a well defined period when the spore-mother-cell has only two chloroplasts, and no longitudinal section of the sporogonium will fail to exhibit a region in which all of the cells are in this condition. As a spore-mother-cell grows older, and increases still further in size, indications of the approaching second division of the chloroplast become manifest.

The second division of the chloroplast repeats exactly the phenomenon of the first division. As a rule both structures concerned are active at the same time, but one may be in a more advanced state of fission than the other. The proportions of the spore-mother-cell at this time are more nearly equal (*fig.* 7) than in the previous condition, *i. e.*, the cell is broader in proportion to its length. As a result the four chloroplasts are distributed around the centrally placed nucleus in such a manner that each occupies the greater part of one of the four tetrahedral divisions of the spore-mother-cell that are present after the separation of the cell contents to form the spores. Sometimes the

second division of the chloroplast follows so closely after the first that the final products are arranged for a short time in the form of a bow around the nucleus (*fig. 9*). Such conditions are however only transitory and result from the elongated forms of such cells.

The structure of the four daughter chloroplasts exhibits this difference from that of the mother structure shown in *figs. 4, 5*. The protoplasmic material that formerly filled the spaces between the starch grains as a spongy network of strands and films now appears to be entirely absent or present only in a much reduced form. The chloroplast has the structure of a vesicle filled with starch grains. It is interesting to contrast this condition with the conditions exhibited in the youngest spore-mother-cells. There the bulk of the chloroplast was made of protoplasm and the few starch grains were insignificant. But in the oldest spore-mother-cells the chloroplast has become one great storage vesicle of starch, conspicuous by the absence of that protoplasmic differentiation which is so characteristic of the chloroplast wherever found.

The problem of the division of the chloroplast and the distribution of the resultant daughter plastids through the cell is full of interest. Does the development of the furrow and the final fission of the chloroplast result from the activity of the protoplasm in or around the structure? It is conceivable that the division has its cause merely in the advantage that might come from a more even distribution of the chlorophyll through the cytoplasm. Thus it is obvious that the arrangement shown in *fig. 5* is very unsymmetrical. Perhaps a strain may be exerted upon a chloroplast bent around the nucleus in such a manner and finally result in its fission, and then lead to a redistribution of the cell contents to bring about a certain balance in the cell. The difficulty of this view as applied to Anthoceros lies in the fact that the number of chloroplasts is fixed at four, corresponding exactly to the number of spores. How could a numerical coincidence so important in phylogeny be left to the play of forces merely acting for symmetry and the advantageous

distribution of chlorophyll throughout the cell? But if we assign the cause of the division to the protoplasm in or around the chloroplast there arise complications very difficult to explain. The four chloroplasts are in the cell long before the nucleus divides. Can it be supposed that cytoplasm would be intrusted with so important a task as the preparation of a chloroplast for each of the four nuclei that are later to preside over the spores before there is any indication that such nuclear division is to take place? The process reverses what would appear to be the natural course of events; *i. e.*, one would suppose that the division of the nucleus would determine the position of the four spores and that the cell contents would arrange themselves later with reference to the nuclei. Perhaps this really is the fact, and it may be merely chance that each of the four nuclei finds a single chloroplast to accompany it in the spore, but such coincidence of numbers would be very curious. A somewhat similar problem is presented by the oospore of Coleochaete, where, according to Oltmanns, eight chloroplasts are formed before the nucleus divides, and then as cell division progresses the nuclei are distributed symmetrically until each becomes associated with one chloroplast.

The structure and behavior of the chloroplast suggests some extremely interesting lines of research. Is there such a substance as plastidplasm, a particular form of protoplasm with morphological characters that may distinguish it from trophoplasm, filarplasm, centrospheres, and other differentiated structures in the cell? If there is a plastidplasm, what form does it assume at the periods of ontogeny, when chlorophyll and other pigments are absent? Is the plastid a permanent organ of the cell, as is perhaps generally supposed? As the plastid is a region of the protoplasm where the pigment is gathered, and consequently the seat of metabolic activity, it is possible that these functions might produce its form and structure. It is even conceivable that the differentiation of a plastid may not lie in peculiarities of protoplasm itself, but represent the outward effects of the metabolic phenomena concerned with its pigment.

It appears to the writer that plant cytology has a very important field open for investigation dealing with the structure of the chromatophore and its place in ontogeny.

We pass now to the second part of the paper, which is to consider the behavior of the nucleus in sporogenesis. The structure of the spore-mother-cell and process of nuclear division in Anthoceros was first described by Strasburger (**'80**, p. 162). The following account entirely supports the essentials of those observations, but the present investigation attempts a more detailed examination of nuclear activities, involving the problems of spindle formation, synapsis, the succession of nuclear divisions, and formation of the walls between the spores.

The fact has already been stated that eight chromosomes are present in the sporophyte, and consequently enter the nucleus of the spore-mother-cell. This nucleus in a resting condition is very similar to the nuclei of the archesporium and shows very little structural differentiation (*fig. 3*). It is small and its linin network is very inconspicuous, but the nucleolus is prominent.

Coincident with the appearance and increase in size of the chloroplast the nucleus enlarges (*figs. 4, 5, 6*) and the threads of linin become very prominent. They are so exceedingly small, however, that it was not possible to determine accurately the structure, even with a Zeiss apochromatic immersion lens under the magnification of 2250 diameters. Dark-staining minute bodies along the linin thread are presumably granules of chromatin, but their distribution could not be ascertained with certainty, nor was it possible to follow the convolutions of the spirem.

The condition of the nucleus after the first division of the chloroplast is shown in *fig. 6*. The spirem thread is strongly developed, but the spotted character of the nucleolus indicates that the latter structure may be the seat of internal changes. Suddenly, as indicated by the rarity of intermediate conditions, the nucleus passes into the state of synapsis. The spirem thread contracts into an irregularly shaped mass that usually lies by the side of the nucleolus. This condition is illustrated

in *fig.* 7. Synapsis in Anthoceros seems to occur after the first division of the chloroplast, and it is very apt to persist through to the second division. The contracted threads of linin then gradually extend outward through the nucleus (*fig.* 8), and finally a spirem condition is attained similar in all outward appearances to that present in the nucleus before synapsis.

The nucleolus after synapsis is often found somewhat fragmented, as is shown in *fig.* 10. Whether or not the nucleolus partly dissolves and contributes material to the linin thread, as has been suggested for certain forms (*e. g.*, Lilium), could not be determined for Anthoceros.

Synapsis in Anthoceros does not appear to the writer to be an artifact, and in this opinion he agrees with the views expressed by a number of investigators who have studied this phenomenon in other types. Particularly favorable opportunities are presented for the solution of this problem in Anthoceros, because one may subject at one time all conditions of the spore-mother-cells to the same fixing fluid. Under these conditions synapsis always appears in that particular region of the sporogonium comprising cells in the condition just before and during the second division of the chloroplast. No nuclei were ever observed in synapsis in other parts of the sporogonium, and nuclei in younger and older cells immediately adjoining the disturbed region presented the typical fully expanded spirem thread. Yet all of these cells had been bathed in the same fixing fluid and experienced identical treatment in the preparation of the slides. Of course it may be claimed that the contents of the nuclei are more subject to shrinkage at the time when synapsis appears, but it should be understood that such a variety of fixing fluids as Flemming's, picro-acetic, corrosive sublimate, chrom-acetic, and Merkel's, all gave identical results. Unfortunately the small size of the nuclear elements in Anthoceros made a detailed study of this interesting process extremely difficult, and led to no definite conclusions as to the meaning of synapsis.

We have seen that events take place during the growth of the spore-mother-cell in the following order: *first*, an increase in

size of the nucleus, the assumption of the spirem condition, and the first division of the chloroplast; *second*, sudden synapsis at the time when there are two chloroplasts in the cell, and often continuing until after the second division of these structures; *third*, gradual emergence of the nucleus from synapsis. These changes present the spore-mother-cell ready for nuclear division with the following arrangement of the cell contents. The four chloroplasts are distributed symmetrically, with the nucleus in the center of the cell. In this condition the cell remains for a period somewhat longer than that of the synapsis, and then prepares for the first division of the nucleus.

It is difficult to recognize the earliest indications that the nucleus is approaching prophase of division. A somewhat later stage is very conspicuous, when the outline of the nucleus becomes angular and a mat of delicate threads surrounds the structure (*fig. 12*). There is a period, however, previous to this condition, when the nuclear membrane appears much less firm and somewhat irregular in outline. This structure follows the spirem stage such as is shown in *fig. 10*, and precedes the unmistakable prophase conditions illustrated by *figs. 12, 13*. Its appearance is given in *fig. 11*, and the following peculiarities should be noted, viz., an irregularly outlined nuclear membrane, a faint linin network, and fragmentary nucleolus. But perhaps the most important characteristics appear outside of the nuclear membrane in the cytoplasm as a delicate web of fibrils closely applied to the nucleus. These fibers are so delicate as almost to defy the reproduction that has been attempted in *fig. 11*. They presage the development of the spindle.

In the prophases the nucleus may exhibit some extremely varied appearances. The form is very irregular, taking on curious wavy angles. Outside of the nucleus there is a region of protoplasm manifestly very different in structure from the neighboring cytoplasm. In overstained preparations this region is very conspicuous, forming a sort of zone around the nucleus, but the details of its structure can only be made out in very favorable preparations. Then it is seen to possess a fibrillar

structure. The delicate threads lie all around the nucleus, but they accumulate conspicuously at the pointed projections (*figs. 12, 13*).

At this time one is apt to find the linin network contracted away from the nuclear membrane and gathered in the form of a confused tangle in the central region somewhere near the nucleolus (*fig. 13*). The chromatic material becomes more prominent and from the ill-defined mass in the interior there emerge four deeply staining chromosomes (*figs. 12, 14*). When the chromosomes are fully differentiated one may expect to find one or two of the angles of the nucleus more prominent than the others, and the fibrils around them taking on the appearance of spindle fibers (*figs. 13, 14*). The process of spindle differentiation is a gradual one, and only in the later stages is it possible to feel sure of the direction that the axis will assume. It is seldom that the two poles appear from the beginning so nearly opposite one another as to have a common axis. It is more usual for the spindle to be somewhat bent at first, as is shown in *fig. 14*. However, ultimately the two poles arrange themselves to form a symmetrical spindle of the form illustrated by *figs. 15, 16, 17, 18*.

The fully developed spindle has an interesting structure, with several clearly marked features well shown by *fig. 18*. The poles are flat, or perhaps slightly convex; they are never pointed. Mantle fibers are conspicuous around the nuclear plate (*figs. 16–18*), but what relation these bear to the chromosomes could not be determined.

In view of the condition reported by Farmer ('94 and '95) for a number of the Hepaticae, notably Pellia, Pallavicinia, and Fossombronia, it became a very important problem to determine whether or not centrospheres were ever present at the poles of the spindles. The writer searched his preparations with great care for these structures, but came to the conclusion that they do not exist in Anthoceros. Sometimes the cytoplasm around the spindle contains large deeply staining granules, such as are shown in *fig. 17*, and these may occupy positions near the ends

of the spindles, but extensive observations would convince anyone that they have nothing whatever to do with centrosomes. The protoplasm in the vicinity of the pole is often very dense in structure, containing no granules and few vacuoles. It frequently presents the appearance shown in *fig. 16*, but it never exhibited indications of that differentiation expected of a centrosphere.

It will be apparent to the reader from this account that the spindle of Anthoceros during the first division follows a history closely parallel to that described for homologous cells in Lilium by Mottier ('97), Hemerocallis by Juel ('97), Equisetum by Osterhout ('97), and Cobæa by Lawson ('98). That is to say, the spindle is organized by numerous delicate fibrils of protoplasm that develop conspicuously during prophase in the cytoplasm around the nuclear membrane. The fibrils are at first somewhat irregularly distributed, but finally become arranged in the form characteristic of the respective spindles.

Strasburger in the third edition of the *Lehrbuch der Botanik*, 1898, p. 67, has introduced the term filarplasm to be applied to protoplasm having the form and activities above described and so clearly established by the researches of his students during the past three years. The writer understands that filarplasm is supposed to be made up of the substance designated by the older term kinoplasm. However, filarplasm has morphological characters, as indicated by its name, and these are the thread or fibril-like structures. The term therefore expresses admirably the facts of morphology without implying or assigning physiological activities to the substance.

It will be very gratifying if future investigation should establish filarplasm as an element in the cells of higher plants differentiated from other forms of protoplasm. This investigation appears to extend the range of mitoses associated with filarplasm into a group of plants much lower than the lowest previously reported (pteridophytes by Osterhout). It is also significant that it should be a class containing one large order, the Jungermanniaceæ, where according to Farmer centrospheres are very marked

features of the mitotic figures. There should be types of plants that will allow us to form some idea of the relationship between filarplasm and the asters and centrospheres present in certain thallophytes, in other words solve the problems of their places in phylogeny. Perhaps the future study of Hepaticæ will throw some light on this question.

The two sets of daughter chromosomes that result from the division at metaphase pass very quickly from the nuclear plate to the poles of the spindle. Here they may frequently be found clustered together in a region of dense protoplasm that surrounds the group, as is shown in *fig. 19*. The development of a nuclear membrane around the chromosomes finally organizes the two daughter nuclei which are connected for a short time by the spindle fibers. These latter structures gradually fade away, beginning at the poles (*fig. 20*), and finally entirely disappear. The daughter nuclei then lie in undifferentiated cytoplasm usually occupying positions between the chloroplasts somewhat as is indicated in *fig. 21*.

The daughter nuclei following the first division pass into a fully developed resting condition. The chromosomes break up into a number of chromatin granules that become distributed over a linin network. A prominent nucleolus appears. In sections of a sporogonium one may always find spore-mother-cells containing two nuclei, although this condition is quickly replaced by the succeeding second division.

The second mitosis in the spore-mother-cell involves both nuclei simultaneously, but the two nuclear figures are developed independently of one another. The two spindles are never united by cross fibers. Sometimes the two spindles will lie almost side by side, but usually they are placed at right angles to each other. Frequently one section of a spore-mother-cell will give a polar view of one spindle and the neighboring section a longitudinal view of the other. An instance of this character has been chosen to illustrate the principal features of their structure and is shown in *fig. 24*. By comparing *fig. 24* with illustrations of the first division (*figs. 16–18*) it will be seen that the spindles of the

second mitosis are much smaller. However they exhibit an essentially similar structure, having flattened poles without centrospheres. Several prominent fibrils make up the center of the spindle which is bordered by an ill-defined set of mantle fibers. The polar view of a spindle shown at the left of *fig. 24* demonstrates that four chromosomes are present at the nuclear plate, the same number that appears during the first mitosis. Prophase and anaphase conditions of the second mitosis are found only with great difficulty and are very unsatisfactory for study because of the small size of the elements involved.

The four nuclei that result from the second mitosis associate themselves each with one chloroplast, and with these become distributed symmetrically through the cell, so that the protoplasm naturally segregates into four regions representing what are later to become the tetrahedral division of the spore-mother-cell (*figs. 26, 27*). The spindle fibers disappear completely.

The problem of the splitting of the chromosomes engaged the writer's attention, but it must be plain that Anthoceros is not a favorable subject for the study of this process. The nearly spherical form of the chromosomes offers immense difficulties in orientation. Farmer ('95) reported some peculiar conditions in the forms studied by him, which he considers as illustrations of the "heterotype" division described by Flemming. They result from the habit that the chromosomes have of doubling on themselves and then being pulled apart as V-shaped daughter chromosomes. The figures are very complex, but Farmer assures us that the division is really longitudinal and not transverse, so that it cannot be interpreted as qualitative. In Anthoceros all evidence that the writer can present indicates that the splitting in both mitoses is longitudinal and after the normal type. Polar views of spindles after the splitting of the nuclear plate sometimes present the daughter chromosomes, eight in number, arranged in four pairs. Such a stage is shown in *fig. 23*, and the grouping certainly indicates that each of the original four chromosomes has divided longitudinally into halves. It is not at all unusual to find the two sets of daughter chromosomes placed as in *fig. 22*, which might

suggest transverse fission, but the writer is not willing to accept this evidence, as such an arrangement would naturally appear when the daughter chromosomes pass away from the nuclear plate to the poles of the spindle.

We have now finished our account of nuclear activities in the spore-mother-cell, but there remains for consideration the description of the manner in which the cell contents are divided to form the spores. After the two mitoses each of the four nuclei lies in a region of dense protoplasm at the side of a chloroplast towards the interior of the cell (*fig. 25*). The bulk of the protoplasm is therefore collected into four masses somewhat apart from one another but connected by very numerous delicate filaments. The cross filaments are very conspicuous but irregular in their arrangement, frequently anastomosing. As shown in *fig. 25* they are not confined to the vicinity of the nuclei but connect all portions of the separated regions of protoplasm. They do not resemble spindle fibers, being much thicker, but have instead the appearance of strands of cytoplasm. Following the condition shown in *fig. 25* one may find stages similar to *fig. 26*. It is plain that the protoplasmic strands have spread sideways and fused with one another so that there is now present a film of protoplasm between tetrahedral regions of the spore-mother-cell. This film marks exactly the position that is finally to be occupied by cell walls when the spores are fully organized.

The peculiarity of the process just described lies chiefly in what seems to be its entire independence of spindle fibers. It is the generally accepted view that the walls crossing spore-mother-cells and pollen-mother-cells are derived from cell plates that result from the fusion of spindle fibers after anaphase. The writer thinks that no investigator has described a condition similar to Anthoceros. The present studies indicate that the spindles of the two successive mitoses completely disappear. This is certainly a very difficult point to determine, but the writer feels confident that the anastomosing strands which connect the four masses of protoplasm in the spore-mother-cell,

as has been shown in *figs. 25, 26*, are formed entirely independently of spindle fibers. How then are the walls formed?

It is certain that the film of protoplasm indicated in *fig. 26* thickens and finally gives place to a straight wall, at first delicate (*fig. 27*) but gradually becoming firmer until its cellulose nature is unmistakable.

The phenomenon is typical of one of the processes recently discussed by Strasburger ('98), in which a cell wall is formed in the interior of the protoplasm. It appears as if the film of protoplasm exhibits the activities present in the "Hautschicht" when it lays down or increases the thickness of a cell wall in the manner known as apposition, involving, at least in part, the change of its own substance into cellulose.

With the separation of its contents the spore-mother-cell as a unit ceases to exist, and a new set of activities begins that may very properly be reserved for discussion in another paper. Some interesting events take place in the spore as it ripens, but technical difficulties interfere greatly with their elucidation.

The peculiar fact that the sporogonium of Anthoceros presents spore-mother-cells in all stages of development makes it possible to contrast the times occupied by the various changes. One cannot establish the actual duration of any process, but within certain limits it is possible to determine the relative periods of each event. It must be assumed that the rate of growth during the season is approximately uniform. The unit of the calculation must be the time necessary to differentiate one spore-mother-cell from the archesporium. As the result of a laborious examination and count the writer ventures to present the following time schedule of the events which take place in the spore-mother-cell. From 12–18 units of time are necessary to bring the spore-mother-cell to the period of the first division of the chloroplast. The division of the chloroplast occupies 6–10 units. The nucleus is in synapsis 12–20 units. For 30–50 units the cell has two chloroplasts. It takes about 25 units after synapsis to produce conditions favorable for mitosis. The first mitosis occupies 1–3 units, and the two daughter nuclei rest for 2–4

units before the second mitosis which is more rapid than the first. There is finally a period of 3–6 units before the spores are definitely organized by the partition walls across the spore-mother-cell.

The results presented by this investigation indicate that the Hepaticæ are likely to furnish some very interesting material for future researches in cytology. From superficial examination it appears probable that the Ricciaceæ and Marchantiaceæ in general present mitoses similar to Anthoceros, but we may hope that some forms exist that will harmonize the peculiarities described for the Jungermanniaceæ with the conditions found in Anthoceros.

EXPERIMENTAL TECHNIQUE.

As stated in the beginning of the paper the writer experimented with a number of fixing agents to determine as precisely as possible their merits or faults. Of these Flemming's formula designated "weak" gave decidedly better results than any other. In Anthoceros, and presumably for other types, the chief test of a fixing fluid is its effect upon the achromatic parts of a nuclear figure. Chromosomes are the least difficult of all the nuclear elements to preserve. Preparations will not infrequently present beautiful views of nuclear plates when the spindles are manifestly in very bad condition. The following is a brief statement of the effects of several fluids upon nuclear figures in the spore-mother-cell.

Chrom-acetic acid fixes filarplasm but the safranin stains diffusely after it, and gentian violet does not hold well in the spindle fibers. If sections fastened to the slide be left several days in weak Flemming the staining qualities with safranin and gentian violet are much improved although it is doubtful if they can be made as good as those presented by Flemming fixed material.

Merkel's fluid (1 per cent. chromic acid 12^{cc}, 1 per cent. platinum chlorid 12^{cc}, water 72^{cc}) even when used for long periods (36 hours) is thoroughly unsatisfactory. The spindles are badly fixed.

Boveri's picro-acetic acid gives beautifully bleached tissue, but achromatic regions are not clearly differentiated although chromatic elements stain well.

Sublimate-acetic (5 per cent. glacial acetic acid in saturated solution of corrosive sublimate) is not good. Nuclear membranes and filarplasm are very poorly preserved.

Hermann's fluid is very much like Flemming's in its effects and is thoroughly satisfactory.

The osmic acid of the Flemming's and Hermann's mixtures appears to give them certain advantages over all other fluids. Although they may not kill and preserve tissue better than some other agents, as for example chrom-acetic acid, certain stains, safranin and gentian violet, differentiate all structures of the cell very much better when they have been used.

SUMMARY.

The number of chromsomes is eight for the sporophyte and four in the gametophyte.

The chloroplast appears rather suddenly in the spore-mother-cell as a differentiated region of the protoplasm, containing several starch grains. When fully developed it has a honeycomb structure, each cavity being occupied by a grain of starch.

The division of the chloroplast is one of simple fission, apparently through forces acting outside of the structure, and perhaps concerned with the film of protoplasm that surrounds it.

Synapsis occurs in the nucleus soon after the first division of the chloroplast. It is not an artifact.

The second division of the chloroplast presents the spore-mother-cell ready for the division of the nucleus. The four chloroplasts, hardly more than vesicles filled with starch grains, are arranged symmetrically in the cytoplasm with the nucleus in the center of the cell.

The resting nucleus has a nucleolus and a spirem thread, which, however, is so small that details of structure could not be determined.

Prophase conditions of the nucleus show the presence of a mesh of delicate fibrils (filarplasm) around the structure. The nuclear membrane is at first irregularly angular, but finally two poles of a spindle are differentiated.

The metaphase presents a spindle with flattened poles, entirely lacking bodies that might be interpreted as centrospheres or centrosomes.

There is a period of rest after the first mitosis when each daughter nucleus has a nucleolus and spirem thread. The two mitoses are, therefore, successive.

The second mitosis presents structural features quite identical with the first.

The chromosomes are four in number in each mitosis. They appear to split longitudinally.

All traces of the spindles become lost soon after each mitosis.

The fully mature spore-mother-cell presents four chloroplasts, each with a single nucleus on the interior side. The protoplasm segregates in these four regions of the cell, leaving spaces that are crossed by many anastomosing strands of cytoplasm. These strands cannot be traced from spindle fibers and appear to have no connection with filarplasm.

The walls separating the cell contents into four spores are derived from films of protoplasm that appear between the chloroplasts with their respective nuclei. The films are formed by the coalescence of strands of cytoplasm that cross the spaces between the four regions of cell contents.

THE UNIVERSITY OF CHICAGO.

LITERATURE CITED.

CAMPBELL ('95) Mosses and ferns.

——— ('98) On the structure and development of Dendroceros. Jour. Linn. Soc. **33** : 467.

FARMER and REEVES ('94) On the occurrence of centrospheres in *Pellia epiphylla* Nees. Ann. Bot. **8** : 219.

FARMER ('94) Studies in Hepaticæ : On *Pallavicinia decipiens* Mitten. Ann. Bot. **8** :35.

——— ('95) Spore formation and karyokinesis in Hepaticæ. Ann. Bot. **9** : 363.

——— ('95) On spore formation and nuclear division in Hepaticæ. Ann. Bot. 9 :469.

JUEL ('97) Die Kerntheilung in den Pollenmutterzellen von *Hemerocallis fulva* und die bei denselben auftretenden Unregelmässigkeiten. Jahrb. f. wiss. Bot. 30 : 51.

LAWSON ('98) Some observations on the development of the karyokinetic spindle in the pollen-mother-cells of *Cobæa scandens* Cav. Proc. Calif. Acad. Sci. Botany 1 : 169.

MOTTIER ('97) Beiträge zur Kenntniss der Kerntheilung in den Pollenmutterzellen einiger Dikotylen und Monokotylen. Jahrb. f. wiss. Bot. 30 : 15.

OSTERHOUT ('97) Ueber Entstehung der karyokinetischen spindle bei Equisetum. Jahrb. f. wiss. Bot. 30: 5.

STRASBURGER ('80) Zellbildung und Zelltheilung.

——— ('98a) Lehrbuch der Botanik, dritte Auflage.

——— ('98b) Die pflanzlichen Zellhäute. Jahrb. f. wiss. Bot. 31: —.

EXPLANATION OF PLATES IX AND X.

All preparations were studied under a Zeiss oil immersion 2^{mm} aper. 1.30 with compensation-oculars. Figures were sketched with an Abbé camera at the following magnifications, *figs. 3–7, 9, 10, 12, 16–21, 24–27*, magnified 1000 diameters; *1, 2, 8, 11, 13, 14, 15. 22*, 1500 diameters and *fig. 23*, 2250 diameters.

All preparations except that shown in *fig. 2* from material fixed in weak Flemming, sectioned 5μ thick and stained on the slide; *figs. 1, 15, 18, 22* and *23* from slides stained with iron-alum-haematoxylin after the method of Haidenhain; all other figures from preparations stained with Flemming's triple stain, safranin, gentian violet, and orange *G*.

PLATE IX.

FIG. 1. Nuclear figures from archesporium, just after the splitting of the chromosomes whose number is large, apparently about sixteen, eight for each daughter nucleus.

FIG. 2. Nuclear figure from antheridium, four chromosomes.

FIG. 3. Spore-mother-cell, early condition of chloroplast with three or four grains of starch.

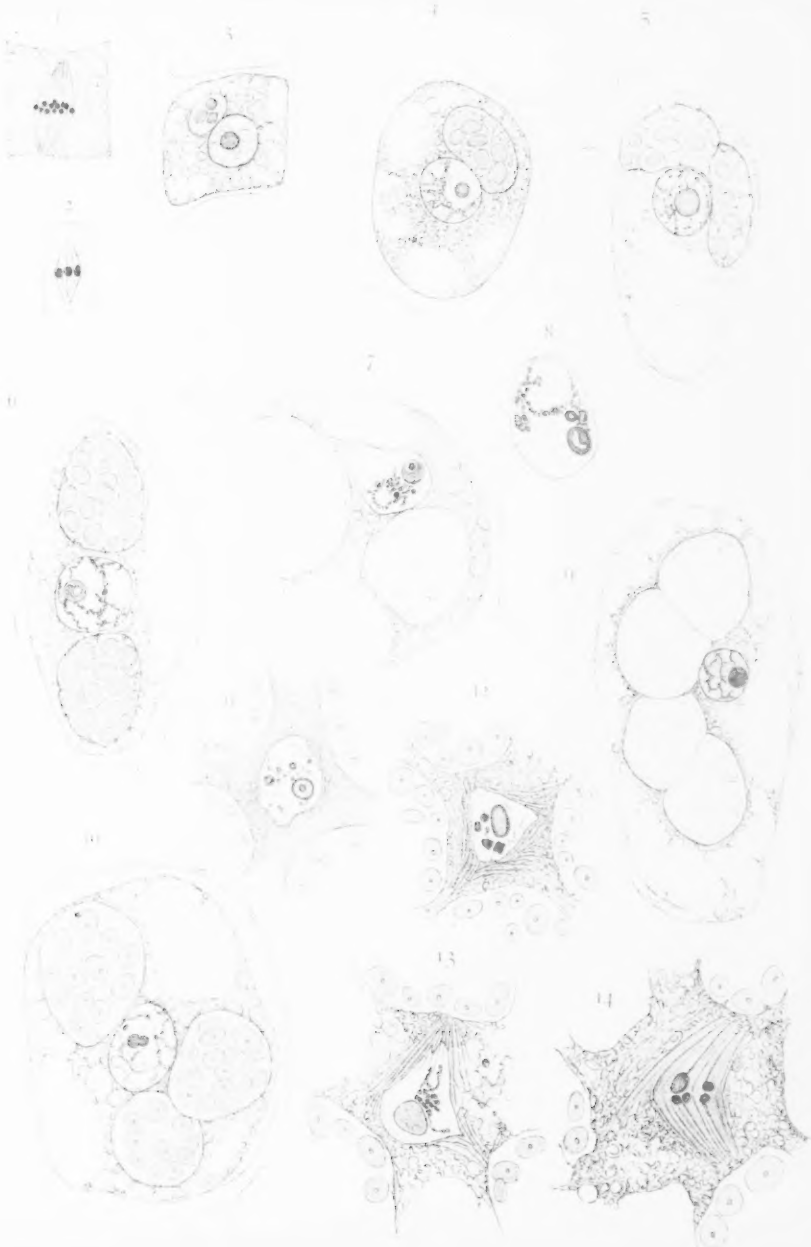
FIG. 4. Spore-mother-cell in more advanced condition than *fig. 3*, linin network more prominent, chloroplast elongating preparatory to division; starch grains prominent.

FIG. 5. First division of chloroplast.

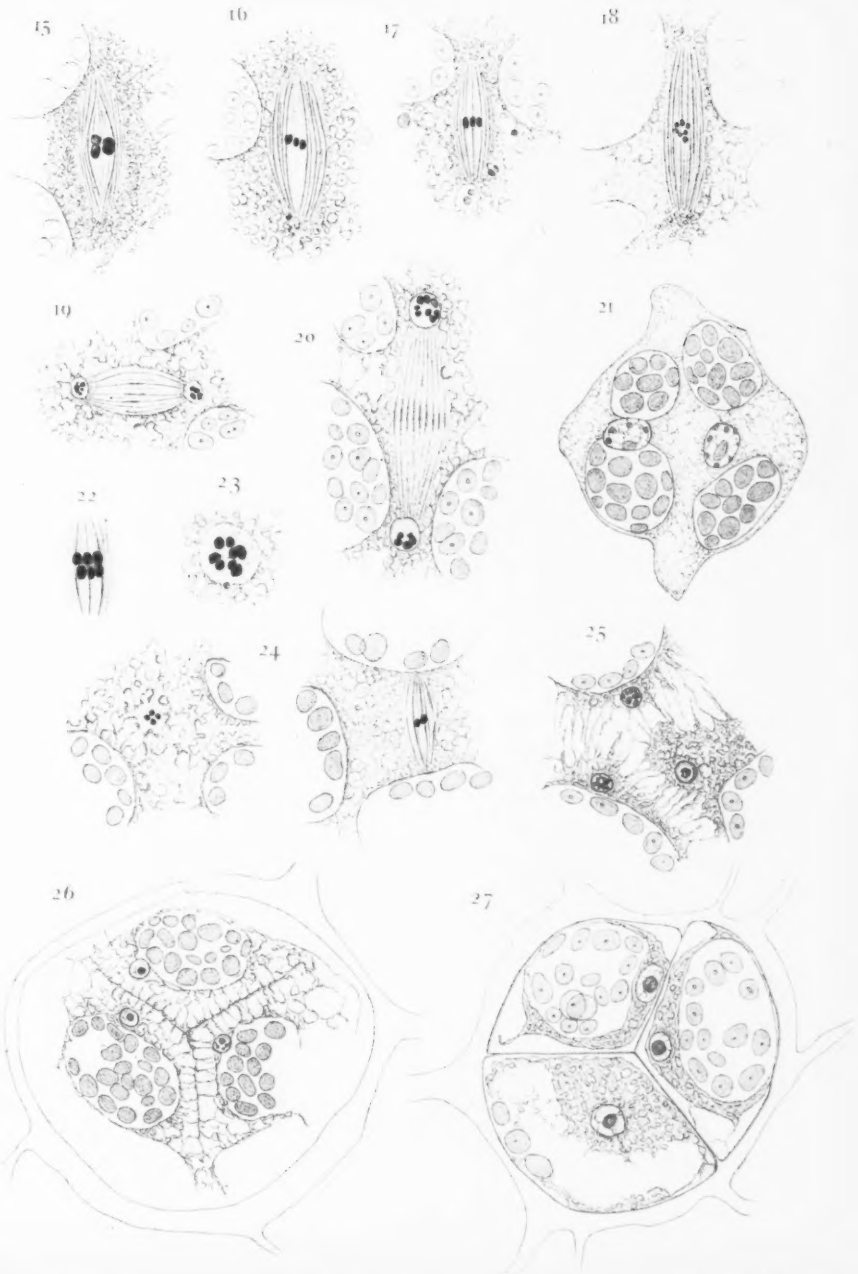
FIG. 6. Two chloroplasts, linin network prominent.

FIG. 7. Synapsis, two chloroplasts.

FIG. 8. Nucleus emerging from synapsis.



DAVIS on ANTHOCEROS.



DAVIS on ANTHOCEROS.

FIG. 9. Second division of the chloroplasts.

FIG. 10. Spore-mother-cell at maturity, four chloroplasts.

FIG. 11. First indication of approaching mitosis, accumulations of filarplasm around the nucleus whose membrane appears less clearly defined.

FIG. 12. Prophase, filarplasm well differentiated.

FIG. 13. Prophase, one pole of spindle developed.

FIG. 14. Late prophase, the two poles of the spindle differentiated, four chromosomes assembled in the equatorial region.

PLATE X.

FIG. 15. Early metaphase, four chromosomes at nuclear plate.

FIG. 16. Metaphase, strongly developed mantle fibers.

FIG. 17. Metaphase, granular cytoplasm around the nuclear figure.

FIG. 18. Late metaphase, after splitting of the chromosomes, very clearly defined spindle.

FIG. 19. Anaphase, spindle still clearly defined.

FIG. 20. Telophase, trace of spindle in equatorial region.

FIG. 21. Spore-mother-cell, daughter nuclei after first mitosis in resting condition, four chloroplasts.

FIG. 22. Nuclear plate at first mitosis, side view.

FIG. 23. Nuclear plate of second mitosis viewed from pole of spindle, indicating longitudinal splitting of the chromosomes.

FIG. 24. Adjacent sections of same spore-mother-cell during second mitosis; nuclear plate of one spindle with four chromosomes viewed from the end; side view of the other spindle.

FIG. 25. Center of spore-mother-cell showing three nuclei, cytoplasm segregated around the nuclei and chloroplasts, delicate strands connecting the regions.

FIG. 26. First indication of the position of the walls that are to be formed in the spore-mother-cell; the coalescence of connecting strands of cytoplasm to form a delicate film.

FIG. 27. Final separation of the protoplasmic contents of the spore-mother-cell.

THE STRUCTURE AND DEVELOPMENT OF CRYPTOMITRIUM TENERUM.

LE ROY ABRAMS.

(WITH SIX FIGURES)

THE genus Cryptomitrium, represented by the single species *C. tenerum* (Hook.) Austin, has not yet been thoroughly studied, and as the plant has been collected in several localities near Stanford University, at the suggestion of Dr. Campbell a study of its structure and development was undertaken, in order, if possible, to determine its relationship to the other Marchantiaceæ.

The first collections were made late in the spring of 1898, at which time the plants were mature, and the spores almost ripe. Some of these plants were placed in alcohol, while others were allowed to become dry, and the earth upon which they were growing was kept until the following September, when the work upon the plant was taken up. A considerable number of the mature sporogonial receptacles were also put up dry, in order to study the germination of the spores.

Some of the pieces of earth with the dried specimens upon them were thoroughly soaked, and then kept well moistened. Within a day or two the tips of the apparently dried-up plants became green and fresh, and in about two weeks the antheridia began to form. All of the material for study was obtained in this way, until after the rains came. Then considerable material was collected from out of doors, where it developed much faster and was healthier than that grown in the laboratory.

THE THALLUS.

Cryptomitrium, like most other Marchantiaceæ, has a flat, dichotomously branched thallus, which in this species is very thin and delicate. The smooth glossy appearance, by which one can easily distinguish the sterile plants from *Fimbriaria*

Californica Hampe, a species almost always associated with it, is due for the most part to the minute stomata.

These stomata are surrounded by eight, occasionally seven, very symmetrically arranged guard cells (*a*, *fig. 1*), and not by five or six, as stated by Stephani.[1] Each opens into a well-developed air chamber (*b*, *fig. 1*), the boundaries or walls of which can be seen easily with a hand lens, forming a fine network under the epidermis.

These air chambers are much the same as in *Fimbriaria Californica*. They are distributed irregularly throughout the green tissue. Only a single layer of cells separates them, and often one cavity is connected with another. Their development begins a little further back from the apical cell than in the above mentioned species as described by Campbell.[2]

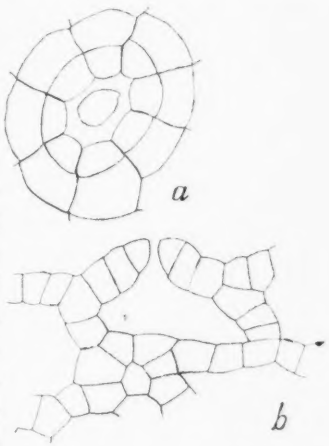


FIG. 1.— Stomata of the thallus. *a*, as seen from the surface. ×600. *b*, transverse section. × 600.

The general appearance and external characters of the ventral scales have been quite thoroughly and accurately described by Stephani (*loc. cit.*), and as their development does not differ from that of other allied genera it need not be repeated here.

As might be expected, both kinds of root hairs, those with smooth thin walls and those with tuberculate walls, which are characteristic of the Marchantiaceæ, are present.

The peculiar oil bodies found in so many of the Hepaticæ were found scattered throughout the thallus, ventral scales, and sporogonial receptacle. The development and composition of these oil bodies found in the Hepaticæ have been thoroughly studied by Pfeffer.[3]

[1] BOT. GAZ. 17: 58–60. 1892.

[2] CAMPBELL, D. H.: Mosses and Ferns, p. 48. 1895.

[3] PFEFFER, W.: Die Oelkörper der Lebermoose. Flora 32: ——. 1874.

THE SEXUAL ORGANS.

Cryptomitrium tenerum is monœcious. The antheridia form a single row just back of the sporogonial receptacle. They are sunk deep in the thallus, and each one is marked on the surface by a small conical ostiolum. These ostiola are very inconspicuous, however, and their presence can scarcely be detected with a hand lens.

The antheridia are developed before the female receptacle, and in Fimbriaria and other allied genera, they are formed on the dorsal side just back of the apical cell (*a*, *fig.* 2). The primary

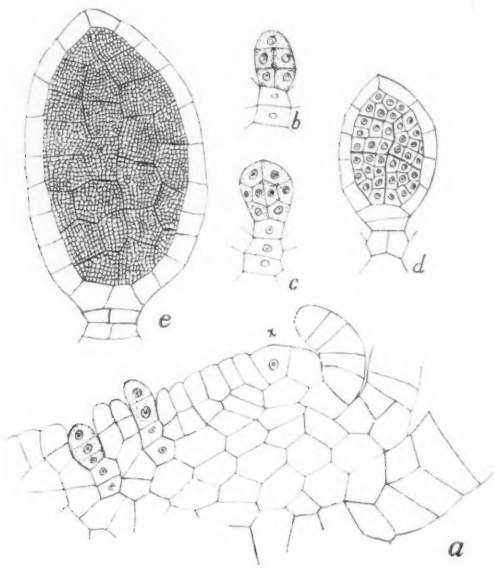


FIG. 2.— Antheridia. *a*, longitudinal section of apex of thallus with two young antheridia; *x*, apical cell. × 600. *b*, *c*, *d*, successive stages of young antheridia. × 600. *e*, full-grown antheridium. × 480.

antheridial cell, when it can first be distinguished from the initial cells around it, is a little larger and stains more deeply than these cells. The first division (*a*, *fig.* 2) is transverse and

divides the primary cell into the stalk or pedicel cell and the antheridial cell proper. The antheridial cell thus formed is then divided into three cells (*a*, *fig. 2*) by two transverse divisions. The next two divisions are longitudinal medial and are at right angles to each other, so that each of the three original cells is divided into four. In many cases, however, only the two lower cells are divided in this way (*b*, *fig. 2*), the top cell remaining undivided for a longer time. The four cells formed from the central cell by these longitudinal divisions are each again divided longitudinally, thus separating the sperm cells from those that go to make up a portion of the antheridial wall (*c*, *fig. 2*). The upper and lower cells take no part in forming the sperm cells, but form, respectively, the upper and lower portions of the antheridial wall. The remainder of the development does not differ materially from that of *Fimbriaria Californica* as described by Campbell.[4] The top of the antheridial wall is not prolonged, however, as in that species, but is only a single row of cells, as in Marchantia. In fact, the full-grown antheridium (*c*, *fig. 2*) resembles very closely that of Marchantia.

The sporogonial receptacle, or carpocephalum, is of Leitgeb's "Compositæ" type.[5] The apical cell of the thallus forms the growing point of the receptacle, but instead of remaining a single cell it divides into two cells. Each of these again divides in like manner. Finally one of the four cells thus formed divides into two, so that there are five growing points. In some cases this last division does not take place, so that there are only four growing points. Five is the usual number, however, for I found only two specimens out of the great number which I examined that had only four growing points. This branch system is somewhat emphasized in the half-grown receptacles, for, at this time, the lobes between the growing points are more developed than the rest, so that the underside of the receptacle has five quite prominent projections or folds. As the receptacle develops, however, these disappear.

[4] CAMPBELL, D. H.: Mosses and Ferns 50, 51. 1895.

[5] LEITGEB: Untersuchungen über die Lebermoose 6: 32.

The dorsal growth of the receptacle is excessive, while the ventral growth is limited to a few layers of cells. Consequently the apical cells (*a*, *fig. 3*) lie very close to the stalk. The lacunæ or airchambers are for the most part confined to a single layer. They are extremely large, however, and are separated

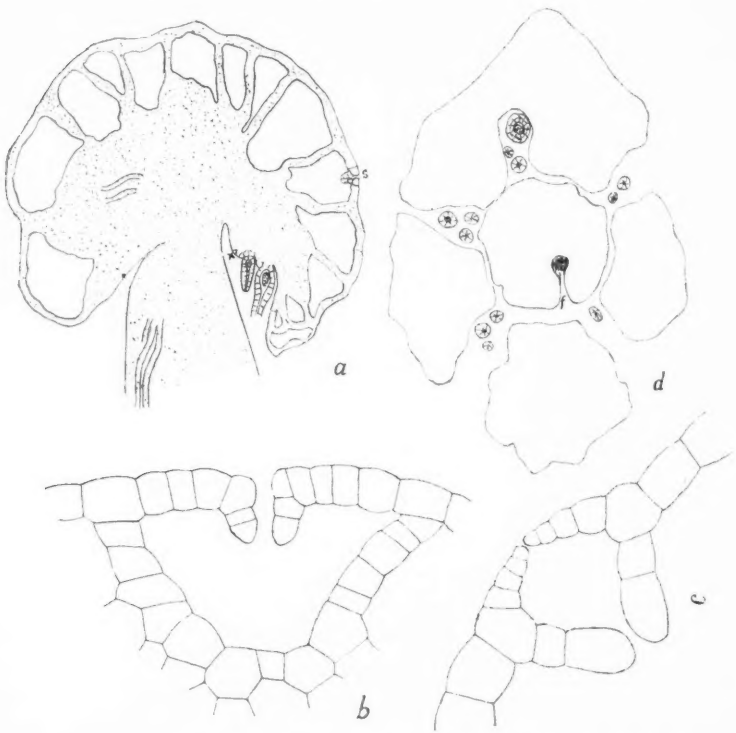


FIG. 3.—*a*, longitudinal section of sporogonial receptacle; *x*, apical cell; *s*, stoma. × 80. *b*, *c*, longitudinal section of stoma. × 600. *d*, transverse section of receptacle; *f*, furrow of peduncle. × 80.

from one another by a single row of cells. Each air-chamber is connected with the exterior by means of well-developed stomata. These peculiar breathing pores are present in several

of the Marchantiaceæ. They are almost cylindrical, and are composed of several rows of cells, which are formed from the original guard cells. These, instead of remaining single, divide by means of inclined walls into several cells. Generally four cells are cut off on the upper side (*b*, *c*, *fig. 3*), and two on the lower side of each original guard cell.

Stephani, *loc. cit.*, reports that there are two furrows on the ventral side of the stalk or peduncle of the receptacle, but I was unable to find such to be the case. In all the specimens examined there was only one. Mr. Howe,[6] also, reports only one furrow in the specimens examined by him. Seen in cross-section (*d*, *fig. 3*) it resembles very closely that of Fimbriaria and Duvalia as figured by Leitgeb.[7] The root hairs, all of which are tuberculate, are found in this furrow. At the base of the receptacle these branch, one branch going to each lobe between the growing points.

The archegonia, although they are on the underside of the receptacle, are in reality on the dorsal side, for they are formed acropetally just back of each apical cell. Hence there are five rows or groups of archegonia (*d*, *fig. 3*). In each of these rows there are usually three or four archegonia.

The development of the archegonium corresponds very closely with that of other Marchantiaceæ. The primary cell becomes much larger than the neighboring cells, and the cell contents becomes much denser, so that it stains very deeply. The first division is transverse. The outer cell forms the archegonium and the inner the stalk. Strasburger[8] states that in Marchantia this outer cell is again divided by a wall parallel to the first, and that the lower cell of the two thus formed forms the foot of the archegonium. Janczewski[9] describes the same thing in *Preissia commutata*. This second division does not take place in Crypto-

[6] Howe, M. A.: Erythea **5**: 87, 88. 1897.

[7] Leitgeb: Untersuchungen über die Lebermoose 6: *Pl. 4, figs. 9, 20.*

[8] Strasburger: Jahrb. f. wiss. Bot. **7**: 416.

[9] Janczewski: Bot. Zeit.— : 418. 1872.

mitrium (*a*, *fig. 4*). Campbell[10] also states that it does not occur in Targionia nor in *Fimbriaria Californica*. The remainder of the development does not differ materially from that of a typical archegonium of any of the Marchantiaceæ, and it need not be repeated here.

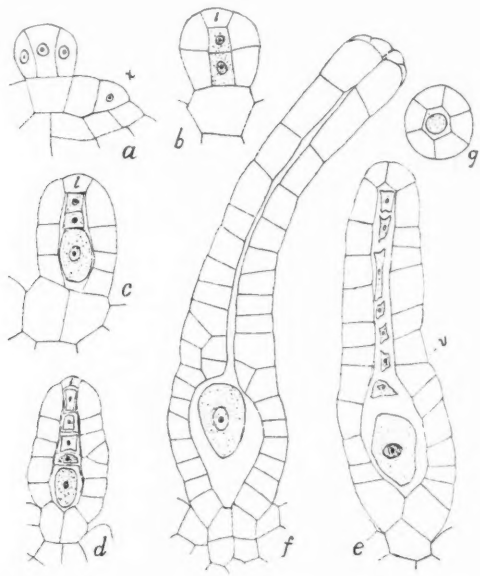


FIG. 4.—Archegonia. *a*, *b*, *c*, *d*, *e*, *f*, longitudinal sections of successive stages; *x*, apical cell; *l*, *l*, *l*, cover cell; *v*, ventral canal cell; *g*, transverse section of neck of an archegonium about the age of *e*. *a*, *b*, *c*, × 600; *d*, *e*, × 480; *f*, *g*, × 300.

The cover cell, which in other forms studied[11] divides into four cells immediately after the neck has been separated from the venter, remains undivided for a considerably longer time in this species (*b*, *c*, *d*, *l*, *fig. 4*). Unfortunately I was unable to determine just when this division took place. Several archegonia were obtained at the age of the one represented in *d*, *fig. 4*, and

[10] CAMPBELL: *op. cit.* 52. 1895.

[11] CAMPBELL: *op. cit.* 30. 1895.

in every case the cover cell still remained undivided. The stages in which the division had taken place (*e*, *fig. 4*) were too old to determine with any degree of accuracy when this division occurred, and it is to be regretted that no intermediate stages were obtained.

Gayet,[12] in a recent article, which has been reviewed by Campbell,[13] states that the archegonia of the Hepaticæ have a distinct apical growth, the same as in the Musci. His conclusions, which are contrary to those of Janczewski,[14] Campbell, and others, are not confirmed by my own observations. While I did not make a very careful study of this point, I could find nothing that indicated an apical cell. The cover cell, which Gayet claims to be the apical cell, does not have the appearance of one. It is much smaller than the upper cells of the neck, and in no way do these cells look as though they had been cut off from it. The fact that the cover cell remains undivided for a considerable time in this species might favor the idea of apical growth, were it not for the fact that the cover cell in this species is even smaller than in other species that have been studied (*d*, *fig. 4*). It looks as if it were lying dormant and not as if it were an active apical cell.

The usual number of neck canal cells is eight, as in the other Marchantiaceæ. In some cases, however (*f*, *fig. 4*), only seven are present.

THE SPOROPHYTE.

Owing to the fact that the embryo in nearly every case lies almost parallel with the stalk, its development has been comparatively easy to make out. One is at once struck with the marked similarity it shows to the embryo of Targionia.[15]

Soon after fertilization the egg cell enlarges to nearly twice its original size. The first division is transverse and divides the enlarged cell into two almost equal cells (*a*, *fig. 5*). The next

[12] GAYET: Ann. des. Sci. Nat. Bot. VIII. —: ——. 1897.

[13] CAMPBELL: BOT. GAZ. **26**: 428–431. 1897.

[14] JANCZEWSKI: *loc. cit.*

[15] CAMPBELL: *op. cit.* 60, 61. 1895.

division is longitudinal (*b*, *fig. 5*). This is followed by another longitudinal division which is at right angles to it. Each of the eight cells thus formed is then divided into two slightly unequal cells by a longitudinal division (*c*, *fig. 5*). The first longitudinal wall is often inclined so that the top cell (*c*, *fig. 5*) resembles very much a two-sided apical cell. The remaining divisions are

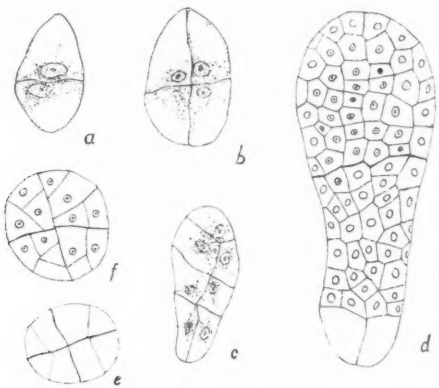


FIG. 5.—Embryo. *a*, *b*, very young stages in longitudinal section. × 600. *c*, somewhat older. × 480. *d*, longitudinal section of still older stage. × 300. *e*, *f*, transverse sections. × 480.

very irregular and are difficult to follow. The young embryo takes on a more and more elongated form. Finally, the central portion almost ceases to grow, so that the embryo becomes dumb-bell-shaped (*a*, *fig. 6*). The upper portion is to form the archesporium, and at about this time, or even earlier (*d*, *fig. 5*), a definite row of cells, which becomes the capsule wall, is formed around the outside. Usually, the first transverse division marks the separation of the capsule and the foot, but in a few cases (*d*, *fig. 5*) this cell remained undivided after the first two longitudinal divisions, so that at the base of the embryo were four large cells.

Soon after the capsule wall is formed the archesporial cells can easily be distinguished, for their protoplasm becomes denser,

and both it and the cell walls, which become very gelatinous, stain deeply (*a*, *b*, *fig. 6*). These gelatinous walls soon dissolve, so that the archesporial cells are set free. Two sorts of cells can easily be made out at this time. The one, the spore

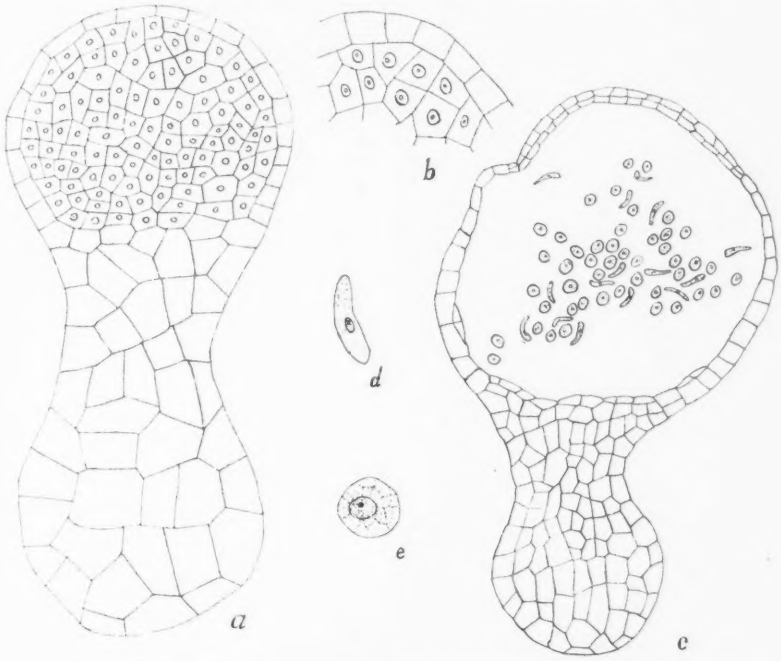


FIG. 6.—Embryo. *a*, longitudinal section of half grown embryo. × 300. *b*, portion of transverse section showing capsule wall and archesporial cells. × 480. *c*, longitudinal section of a nearly mature sporogonium, showing spore-mother-cells and young elaters; *o*, operculum. × 150. *d*, young elater. × 600. *e*, spore-mother-cells. × 600.

mother cells, are almost spherical. Their nuclei are large and distinct (*e*, *fig. 6*), and are surrounded by closely reticulated protoplasm. The other, the young elater cells (*d*, *fig. 6*), are elongated. Their nuclei, though quite distinct, are much smaller than those of the spore-mother-cells.

The foot of the sporogonium (*c*, *fig. 6*) is not so well developed as in Targionia, being not more than one third as large as the capsule, which is large and globular. At maturity the capsule is regularly dehiscent at its apex by an operculum. This operculum, as stated by Howe (*loc. cit.*), is composed of two rows of cells (*o*, *fig. 6*), while the remainder of the capsule wall is, for the most part, only one cell thick. Near its base, however, an apparently continuous ring, composed of only two rows of very small cells occurred in all the specimens examined.

The ripe spores germinate very slowly. During the months of October, November, and December, several cultures were made of spores which had ripened in the previous April, and in no case did they germinate until eighteen or twenty days after they were sown. Their germination and manner of growth correspond very closely to that of Targionia.

SUMMARY.

In comparing *Cryptomitrium tenerum* with the other Marchantiaceæ, I found, as Stephani claimed, that it was undoubtedly very closely related to Duvalia. I had no specimens of Duvalia, however, and was dependent upon Leitgeb's[16] description. Both genera are monœcious. Both have the same minute stomata surrounded by seven or eight very symmetrically arranged guard cells. Stephani states that Cryptomitrium has two furrows in the peduncle, while Duvalia has only one; but only one furrow was present in the specimens I examined, so that neither this difference between the genera, nor the difference in number of guard cells that Stephani described, exists.

The receptacle of Duvalia is nearly spherical, while that of Cryptomitrium is disk shape. In other respects the receptacles resemble each other very much externally; but in the development of the receptacles there is a great difference. Duvalia, according to Leitgeb's account, belongs to the type of the Marchantiaceæ, which has the growing point of the receptacle at its

[16] LEITGEB: *op. cit.* 87–90.

forward margin. In Cryptomitrium the receptacle is a branch system, such as Leitgeb attributes to Marchantia and one or two other genera. This fact alone would be enough to outweigh all the minor external characters referred to, were it not for the fact that, although Leitgeb puts Fimbriaria in the same type as Duvalia, Campbell (*op. cit.*) found that the receptacle of *Fimbriaria Californica* belongs to the "Compositæ," or branching type.

While one hesitates to criticise the classical work of Leitgeb, the quality and accuracy of which is for the most part remarkable, it does not seem reasonable that plants resembling each other as closely as Cryptomitrium and Duvalia should differ so much in respect to the growth of their receptacles, to say nothing of the fact that species of the same genus, Fimbriaria, should also have this difference. It would seem more probable that Leitgeb was mistaken. Probably if one should carefully examine Duvalia and also the species of Fimbriaria which were studied by Leitgeb, it would be found that these too have as many growing points as there are groups of archegonia. Should this not be the case the apparent close relation between Duvalia and Cryptomitrium would be only apparent, and the latter would then, perhaps, have to be considered more nearly related to Marchantia, and *Fimbriaria Californica* could then no longer be considered as a Fimbriaria, for the difference in the two kinds of receptacles is too great to occur within the same genus.

LELAND STANFORD JR. UNIVERSITY.

BRIEFER ARTICLES.

NOTES OF TRAVEL. I.

VENEZUELA.

THE aim of the expedition with which the writer is connected, as planned by Mr. Barbour Lathrop, of Chicago, and carried out at his own expense, has forbidden any exhaustive research into the botanical resources of South American countries. It has permitted rapid comparisons, however, and it is these comparative sketches which it is believed will interest American botanists.

The first approach to a great continent, if it has thousands of square miles of unexplored territory in it, as South America has, is always impressive. La Guayra, the principal port of Venezuela, satisfies one's preconceived notions of tropical luxuriance of vegetation. The steep mountains behind the town shut it in like a green wall, and the low hanging clouds and dark rainy valleys, into one of which the famous railroad to Caracas disappears, are characteristically tropical. By characteristically tropical the writer may give a wrong impression, since what could be characterized as tropical in one region might not be true of another. The xerophytes are as abundant in the tropics as in temperate regions, although in the popular mind they are not characteristic of the tropics. Venezuela landscapes show a larger proportion of xerophytes than I had expected to see, and a ten minutes' tram ride to the small bathing place of Moquendo gave me a good opportunity of seeing the characteristic cactus vegetation of the coast. Almost barren patches of reddish-brown soil and frequent signs of prairie fires on the hillside surprise one, while the tufted grasses, agaves, and cacti give the whole a decidedly arid look. The climate of La Guayra is a dangerous one for foreigners, as the malarial fevers there are very severe. We were informed, however, by intelligent English people living in Caracas that the latter are no more severe than those of Caracas itself. From my friend's most uncomfortable experience it is evident that the capital has a serious form of malarial fever, and great care must be exercised to avoid exposure after sunset.

The La Guayra and Caracas railway has some of the most picturesque scenery in the world. Twenty-three miles of track are necessary to cover seven as the crow flies, and the curves and zigzags along the coast give glimpses of great grandeur. The disappointing part of the landscape lies in barren soil and unmistakable signs of aridity. Curious cereuses and acacias or Prosopis, and a gigantic species of Asclepias with flowers three times the size of our *A. Cornuti* attract one's attention, while the fine-leaved forest trees in the valleys give the landscape a much more northern aspect than would be expected. Two views strike the traveler most favorably; one from a curve in the road which overlooks the coast, where, spread out below, are plantations of sugar cane and banana, fringed with the most graceful of cocoanut palms that stand out like dark green plumes against the white surf; and the second, some distance nearer Caracas, where the road crosses a ravine which drops into a narrow valley, 1500 feet deep and completely clothed with forest.

The vegetation effect, while most impressive, is not truly luxuriant, and unmistakable signs of aridity are everywhere present. Curious arid ridges and isolated peaks along the sides of scantily covered valleys give the impression of poor soil and rapid erosion. There is a rumor that these arid patches were once wooded, but that injudicious removal, constant forest fires, and later prairie fires have denuded them. Dr. Ernst, the Venezuelan botanical authority in Caracas, does not believe this. He declares they have been barren from prehistoric times.

Caracas lies 2632 feet above the sea, surrounded by barren hillsides whose summits are covered with dense forest.

There is very little of botanical interest in Caracas itself. A few interesting private gardens lie across the small stream which flows through the town. One in particular contained a number of curious cultivated plants. In it was the most remarkable fountain I have ever seen. From a hideous cement imitation of a boa constrictor spouted a small stream of water. Around, on little artificial islands, were statues of storks in the act of swallowing. Growing on these islands were large guava trees, oranges in fruit, and tufts of Egyptian papyrus, while for a border to the basin a row of blossoming strawberry plants and Chinese hibiscus bushes had been planted.

The coffee estate of Señor R. Dalla Costa Mosquera, one of the few within the city limits, is well worth a visit and has in it a magnifi-

cent avenue of St. Domingo mahogany trees. It illustrates the coffee culture of Venezuela very well, which is in marked contrast to that of Brazil by its employment of shade trees. These cast a relatively deep shade over the whole plantation, and gave the impression of a thickly planted grove. In Brazil no shade trees are employed, and in Ceylon and Java they are planted very sparsely among the coffee trees. I seriously questioned the advisability of such heavy shading and was informed simply that it was considered advantageous. Some of the best coffee in the world is grown in Venezuela, but very little of it reaches the American market because the latter demands principally the cheaper Brazilian sorts. On the best Venezuelan estates the method of pulping the coffee berry before drying is in use, while the majority of Brazilian coffee growers still cling to the old method of drying the berry first and removing the dried pulp afterward. There are large Brazilian estates where this method has been given up and the best machinery is in use.

Nothing was seen in Caracas of the cocoa industry, although some of the finest cocoa in the world is grown along the Venezuelan seacoast and in the interior about Maracaibo.

In every city one of the most interesting places for a botanist is the market, and in Caracas it is characterized by an extraordinary show of flowers. Tuberoses, white double violets, delicate purple irises, Easter lilies, and curious bouquets made up of double columbines, marigolds and lilies surrounded with tissue-paper lace were most in favor at carnival time. Curious pear-shaped, thick-skinned shaddocks which are used for preserve making; long, chocolate-brown, melon-like squashes, with orange-yellow flesh; immense green watermelons with squash-like meat, showing how careless the growers are about the interbreeding of their squashes and melons; bright yellow "curcumas" with orange-colored, mealy flesh, related to the curcumas of Peru, which are used in the manufacture of a favorite ice; peaches and apples of inferior quality grown in the mountains of the interior; together with the usual number of vegetables of quite inferior varieties compose the piles of produce on the well regulated stalls. This part of the market is much more appetizing than the other section, where the most disgusting looking strings of salted meat are hung for sale.

There is in Caracas a National Society of Agriculture which aims to diffuse intelligence regarding the culture of agricultural products,

to teach the use of fertilizers and soiling crops, etc. But it is governmental, and when that is said of almost any concern in South America, it means that it is subject to rapid political changes.

The courtesy of Mr. Alamo, Assistant Secretary of Agriculture and Mr. Romero, secretary of the society, could scarcely have been greater.

To any foreign scientist, Dr. A. Ernst, the professor of botany in the University, is an invaluable acquaintance. His long experience with Venezuelan conditions and his fund of information on the botanical resources of the country are the result of a wide acquaintance and numerous expeditions into the interior. His advanced years make it impossible for him to continue his work as a collector, but his vigorous mind and excellent memory make his suggestions most valuable to a traveling scientist.

As a young German botanist he came to Venezuela on a collecting trip and was invited by the government to remain. Revolution has followed revolution, but, through his refusal to meddle in the politics of the country in the first place, and his undeniable ability as a scientist in the second, he has kept the position he now holds as one of the most profound scientists and highly esteemed citizens of the republic. His years of labor in an attempt to educate Venezuelan youth to an appreciation of botany have not, it must be regretted, left him hopeful of the final outcome, and from what the writer could learn there is nothing to encourage foreign scientists to engage in governmental work in Venezuela.

Two railways lead from Caracas into the interior, one English, the other German ; neither, however, takes the traveler anywhere near the most interesting region of the Sierra Nevada, which lies, by mule or horseback, some four to five days travel from Caracas. In this Sierra Nevada range are snow-capped peaks and a vegetation that is said to be most luxuriant and peculiar. For the botanical explorer this interior mountain region will prove rich in new forms. Mr. André of Trinidad, a well-known orchid collector, informs me, however, that orchids are more abundant near the coast in the neighborhood of Cumana. In order to explore these interior regions the traveler must have at least a fair knowledge of the Spanish language and should arrange his baggage in square leather boxes not over two feet in largest dimension, suitable for donkey or mule transport. The food obtainable will be of the very poorest quality ; and, as the *peones* live in huts

of most unsanitary character, great care will be necessary to avoid contracting the numerous diseases associated with such conditions.

Caracas is not a favorable place from which to explore the resources of Venezuela. It lies too far from the most interesting portions of the country. The Orinoco can be ascended better from Trinidad, and the Sierra Nevada requires an expedition on muleback to reach it. From descriptions given by travelers on the Orinoco and its branches, the dangers from fever in the forest regions of Venezuela are very great, and anyone undertaking their exploration risks his life. Mr. E. André, whose travels into the interior have been as extensive as any of recent years, said he would not think of taking with him any person who had not lived at least two years in the tropics and become acclimated as far as possible to conditions similar to those in Venezuela.—DAVID G. FAIRCHILD, *U. S. Department of Agriculture.*

SOME SPECIES OF TETRANEURIS AND ITS ALLIES.

WE sometimes hear the statement that the difficulties for the systematic botanist are being multiplied by the breaking up of so many of the old genera and the creation of new species from former aggregates, but practical experience shows, it seems to me, that segregation, when based on describable characters, certainly simplifies. The replacing of the untenable *Actinella* by *Tetraneuris*, *Rydbergia*, and *Picradenia* (Pitt. **3**: 265), is a case in point.

The reduction of several good species to one (an aggregate) makes necessary a description so general that the amateur in the field has no difficulty in placing the most aberrant form until he collects a suite of specimens clearly unlike. In the past, reduction of species has often occurred because certain ones were rare and hence not well represented in the herbaria, but it seems unfair to eliminate a species simply because it exists in a locality not easily accessible or rarely visited.

Being located in the center of distribution of *Tetraneuris* and its allies, I became interested in the group. The following notes and descriptions are offered as supplementary to Dr. Greene's valuable paper cited above.

TETRANEURIS ACAULIS (Pursh) Greene, Pitt. **3**: 265. 1898.

Galardia acaulis Pursh, Fl. **2**: 743. 1814. *Actinella acaulis* Nutt. T. & G. Fl. **2**: 381. 1842, etc.

As limited by the earlier writers this is a variable but a recognizable species. When many of the following were incorporated, the difficulties of the field botanist were multiplied several fold in respect to this species.

TETRANEURIS ACAULIS **cæspitosa**, n. var.—Strongly matted, depressed-spreading, the numerous branches of the caudex much thickened by the imbricated leaf-bases: leaves very numerous and crowded, densely silky-lanate as are also the scapes and involucre: heads nearly sessile or on scapes 3–6^{cm} long.

That specimens of this variety exist in some herbaria as *T. acaulis* is possible, though in the large series in the Herb. Mo. Bot. Garden none were found. Its matted habit, silky-lanate leaves and very short scapes easily separate it. It occurs sparingly on sandy ridges in the foothills. Laramie hills nos. 1890 and 4314 represent it.

Tetraneuris simplex, n. sp.—Tap root vertical, short, comparatively small with few or many secondary roots: caudex short, consisting of one or more thick crowns which are densely covered with brown dead leaf-bases: leaves appressed-pubescent (not silky), nearly naked in the axils or sparsely long-hairy, glabrate in age when the fine punctation becomes evident, crowded on the crowns, ascending or erect, linear-spatulate, tapering only slightly to the margined base, subacute, 4–7^{cm} long: scapes simple, single from the crowns, 15–25^{cm} high, slender, erect, lightly pubescent below, becoming silky or lanate above and on the involucre: head large, 2.5–4^{cm} across; rays with a broad ligule (5–8^{mm}): akene pubescent.

To include this with *T. acaulis* is not conducive to clearness. Dr. Greene (*l. c.*) has separated some of the other well marked species and this seems to me to be as good as the best of them. I have had this and *T. acaulis* under observation for several years, and there is not the slightest possibility of confounding the two in the field. *T. acaulis* is always cespitose, often in considerable mats, the scapes are shorter, the heads smaller, and the rays narrower than in *T. simplex*. The leaves of the latter are comparatively glabrate from the first, strongly in contrast to the silky or even lanate pubescence of the other.

That collectors have never been inclined to call this *T. acaulis* is shown by the fact that specimens of it occur in the herbaria just as often ticketed *T. scaposa* or *T. scaposa linearis*, though to these it is not so closely related. Whether *T. acaulis* or *T. simplex* is the original *Galardia acaulis* is difficult to determine, but the term "pilosa" in the original description, and the

agreement in Nutt. Gen. 173 and T. & G. Fl. that the leaves are "sericeously (silky) villous," and that the plants are aggregated in dense tufts, suggests the separation that is now proposed.

Besides a large series of plants from near Laramie, specimens of *T. simplex* have been examined as follows: T. A. Williams, Pine Ridge, Neb.; J. Schenck, Neb., 1893; H. J. Webber, Pine Ridge, Neb., 1889; Henry Engelmann, North Fork of the Platte, 1858; A. S. Hitchcock, Kan., 1895, no. 289; C. H. Thompson, Kan., 1893, no. 169; Capt. Bryan's Expedition, Lower Pole Creek, Wyo., 1858; R. S. Williams, Great Falls, Mont., 1891, no. 82; G. E. Osterhout, Livermore, Colo., 1898; M. E. Jones, Cheyenne Cañon, Colo., 1878; Hall & Harbour, no. 275.

Tetraneuris incana, n. sp.—Root rather slender, simple or branched: caudex simple or few branched, the crowns enlarged by a dense covering of the broadly expanded bases of the petioles which are more or less involved in white, hirsute wool: leaves crowded on the crowns, silvery-white with an appressed pubescence, linear-oblanceolate, 2–4^{cm} long: scapes naked, single from the crowns, slender, curved-ascending, 1–2^{dm} high, the fine silvery pubescence slightly spreading: involucre silvery-silky, bracts few, shorter than the 1^{cm} high disk, the outer oblong, obtuse, the inner spatulate, scarious margined: rays few, the ligule as long as the disk: disk corollas sprinkled with resinous globules and toward the summit strongly thickened by a dense, penicillate, glandular beard: pappus scales oblong, aristate: akene slender, nearly as long as the corolla, pubescent.

This rare species is strongly marked in its close, silvery pubescense, its nearly simple caudex, its silvery involucre, and its dense coat of glandular hairs on the corollas. The only collections of it at hand are no. 393, Platte hills, near Fairbanks, July 11, 1894; no. 5006 (type number), by Mr. Elias Nelson, Wallace creek, July 30, 1898; and a specimen by Mrs. Muth, Lewis & Clarke co., Mont. Its habitat is white clay ridges among the barren hills.

TETRANEURIS TORREYANA (Nutt.) Greene, l. c.

Actinella Torreyana Nutt., Trans. Am. Phil. Soc. 7: 379. 1841.

A strong species of the central-eastern Rocky mountains; in several forms, but always tufted, strongly punctate and nearly glabrous except on the caudex; somewhat variable as to the width and rigidity of the leaves. My no. 4810, from the Platte hills near Fort Steele, June 18, 1898, are nearly typical; nos. 4571 and 4747, June 1898, from the Tertiary clays of south-central Wyoming have broader, laxer leaves

than the original description permits. A form represented by no. 4327 and some earlier collections, from the limestone ledges of the Laramie hills, is strongly matted and has the branches of the caudex enormously thickened and protected by the densely lanate leaf-bases. Add to this its large root, broad green leaves, and the copious secretion of its punctate glands, and it might well stand as var. *glandulosa*. It is in this species that the salient character of the genus (4-nerved ligules) often fails; 5–8 nerves are not infrequent.

Tetraneuris Mancosensis, n. sp.—Tufted, with woody root and multicipital caudex, the short thickened crowns clothed with the expanded, membranous, lanate leaf-bases: leaves glabrous, 4–8^{cm} long, crowded on the crowns, linear or linear-oblanceolate, acute or cuspidate, rather minutely punctate: stems few to several, bearing a few (usually 2) distant leaves, 2^{dm} in length (including the long monocephalous peduncle): heads large, disk about 1^{cm} high; involucre silky-lanate, the bracts in two or three rows, the inner oblong or somewhat expanded upwards by the scarious margins: paleæ of the pappus oblong-elliptic with an acumination as long as the body proper, equaling the disk corollas: ligules of the rays 15–18^{mm} long, 6–8^{mm} broad.

Collected by Professor C. S. Crandall, Mancos, Colo., June 29, 1898, and distributed as *Actinella scaposa linearis* Nutt. It is in fact, however, much nearer to *T. Torreyana*, from which its slenderer, longer, and less conspicuously punctate leaves, its nearly glabrous two- or three-leaved stems, its long-peduncled heads, and long pappus paleæ at once separate it.

TETRANEURIS LANATA (Nutt.) Greene, l. c.

Actinella lanata Nutt. l. c.

A rare plant of the arid interior, on dry ridges on the high plains. Only the following collections of it have been secured by the writer: 3068 and 4726 from Green river, in 1897 and 1898 respectively; 4607 from Ft. Bridger, June 9, 1898 are young specimens, but probably the same. More or less of the wool is permanent even on the leaves, though these show some punctation on the glabrate areas.

RYDBERGIA GRANDIFLORA (T. & G.) Greene, l. c.

Actinella grandiflora T. & G., Journ. Bost. Soc. Nat. Hist. **5**: 110. 1847.

This occurs in abundance in the alpine regions of all our mountain ranges.

PICRADENIA RICHARDSONII Hook., Fl. **1**: 317. *pl. 108.* 1833.—That authentic specimens of this occur in this range is possible, but it seems

quite certain that the wide range attributed to this species in Gray's *Synoptical Flora* is due to other species being included. With the erection of *P. floribunda* (Gray) Greene and *P. canescens* (Eaton) Greene into species the area covered has also been segregated. But even after the establishment of the two following species some Rocky mountain forms, such as my no. ——, are still left to represent the original species.

PICRADENIA LIGULÆFLORA Aven Nelson, Bull. Torr. Bot. Club **25**: 378. 1898.

This species is proving to be far more common than was at first suspected. As to habitat see notes on the following species.

Picradenia macrantha, n. sp.—Caudex branched, each branch surmounted by a few to several crowns; crowns clothed with the lanate leaf-bases: stems single from each crown, erect, fascicled, sparsely pubescent, somewhat striate, about 15^{cm} high: leaves glabrate, not conspicuously punctate, rather numerous on both crowns and stems, slender petioled, the blade variously parted into linear divisions, most of them pedately trifid, some of the stem leaves pinnately parted or twice trifid, the uppermost sometimes simple: heads large, peduncled, one to five on each stem (generally two or three): involucre shorter than the disk flowers, outer bracts lanceolate, nearly glabrous, united for half their length, 6^{mm} long; inner oblong, acute, scarious margined: rays 6–10, chrome yellow, the ligule $15–18^{mm}$ long and one third as broad: pappus scales 5–7, lanceolate, shorter than the corolla.

Allied to *P. ligulæflora,* from which its larger size, conspicuous rays, longer peduncles, fewer and less resinous heads serve to separate it. The habitat of this is open, stony slopes in the mountains or hills, while *P. ligulæflora* occurs on dry, clayey, alkaline ridges or flats on the open plains. Type number 4830, Fort Steele, June 18, 1898. What seems to be the same thing is no. 1688, Centennial hills, Aug., 1895; also South Park, Colo., 1878, by Marcus E. Jones.—AVEN NELSON, *University of Wyoming.*

PYCNANTHEMUM VERTICILLATUM, A MISINTERPRETED MINT.

MR. W. W. EGGLESTON has called my attention to a Pycnanthemum, abundant about Rutland, Vermont, which has been placed by recent botanists with both *P. muticum* Pers. and *P. Torreyi* Benth., but appears different from either of those species. In its habit

the plant matches the original plate published by Michaux[1] with his description of *Brachystemum verticillatum*. Fortunately a portion of Michaux's type specimen is preserved in the Gray Herbarium, and a careful comparison with that leaves no doubt that Mr. Eggleston's plant is the same. Another Vermont specimen collected by Robbins, and a number of specimens from more southern states possess the same characters as the Michaux plant, and with it represent a species fairly distinct from either *P. muticum* or *P. Torreyi*.

Brachystemum verticillatum was described by Michaux from the mountains of Pennsylvania and Carolina. It was soon transferred to Pycnanthemum by Persoon,[2] who merely accepted Michaux's description without comment. Pursh in his *Flora*, took up the name, modifying the description slightly, but Pursh's plant is said by Bentham, in his monograph,[3] to be the same as *Pycnanthemum aristatum* Michx. In the same work[4] Bentham placed *P. verticillatum* under *P. lanceolatum* Pursh; but subsequently, in De Candolle's *Prodromus*,[5] he reinstated it as a doubtful species. In the *Synoptical Flora* Dr. Gray treated the plant as a form of *P. muticum* Pers.

In 1891 Otto Kuntze[6] took up the obsolete[7] generic name *Koellia* Moench,[8] and transferred to it Michaux's *Brachystemum verticillatum*, though from his statement it is apparent that he meant *Brachystemum muticum*, and was merely following Dr. Gray in regarding that and *B. verticillatum* as the same. In 1894, in their list of the plants of southwestern Virginia, Dr. Small and Miss Vail treated the plants as different species, Dr. Britton[9] there transferring *Brachystemum muticum* Michx. to Koellia. *K. Torreyi* Kuntze (*Pycnanthemum Torreyi* Benth.) was also then recognized as a distinct species. In the *Botan-*

[1] MICHX., Fl. **2**: 6. *pl. 31*.

[2] PERS. Syn. **2**: 128.

[3] BENTH. Lab. 328.

[4] BENTH. *l. c.* 330.

[5] DC. Prodr. **12**: 190.

[6] O. K. Rev. Gen. **2**: 520.

[7] There is little reason to suppose that the name *Koellia* will be generally accepted among botanists. Though that name is printed instead of Pycnanthemum in Engler and Prantl's *Natürlichen Pflanzenfamilien*, Professor Urban of Berlin writes that, not having been used within fifty years of the time of its original publication, it must be rejected by the "Berlin rule"; and that only through an unfortunate oversight was Pycnanthemum not taken up in its stead in the preparation of the *Supplement* to Engler and Prantl's work.

[8] MOENCH Meth. 407.

[9] BRITTON in Small and Vail, Mem. Torr. Bot. Cl. **4**: 145.

ical Club Check List,[10] however, *P. Torreyi* Benth. is reduced to *K. verticillata*. Thus within three years this plant, which was quite obscure to the great monographer of the Labiatæ, under the inspiring influence of a resuscitated generic name was combined with at least two other species: Otto Kuntze, following Dr. Gray, considered the plant a form of *Pycnanthemum muticum*; then it was treated as a species distinct from both *P. muticum* and *P. Torreyi;* then during the same year it was united with *P. Torreyi*.

As stated above, a study of the type of *Brachystemum verticillatum* Michx. shows that plant to be fairly distinct from either his *B. muticum* or the later *Pycnanthemum Torreyi* Benth. The following description and notes may help to distinguish this plant, and show the points of similarity and of difference between it and those with which it has been associated.

PYCNANTHEMUM VERTICILLATUM Pers.—Stem shortly and rather closely pubescent especially above, the branches loosely subcorymbose: leaves ovate-lanceolate or lanceolate, entire or slightly toothed, subsessile, mostly glabrous; the reduced upper ones subtending the rather dense terminal or verticillastrate glomerules, densely short-pubescent: bracts of the glomerules ovate-lanceolate with subulate tips, more or less pubescent on the backs, and with ciliate margins: calyx 5·5^{mm} long, with the 5 hispid lance-subulate teeth about equal: corolla 7 or 8^{mm} long, the upper lip short-oblong or obovate, the lower with the oblong middle lobe twice exceeding the lateral ones: stamens included, scarcely 1^{mm} long.—Syn. **2**: 128; Benth. in DC. Prodr. **12**: 190; Gray, Am. Jour. Sc. **42**: 47. *P. lanceolatum* Benth. Lab. 330, in part, not Pursh. *P. muticum* Gray, Syn. Fl. **2**: 355, in part, not Pers. *Brachystemum verticillatum* Michx. Fl. **2**: 6. *pl. 31*. *Koellia verticillata* Kuntze, Rev. Gen. **2**: 520 (as to synonym only); Small and Vail, Mem. Torr. Bot. Cl. **4**: 146 (at least as to Farmer Mt. specimen; but not as to note, which apparently refers to *Pycnanthemum clinopodioides* Gray); Britton, Mem. Torr. Bot. Cl. **5**: 280 (excluding syn. *Pycnanthemum Torreyi* Benth.).—Specimens examined: Pennsylvania (*Michaux*); Colchester, Vermont (*Robbins*); Rutland, Royalton and Clarendon Springs, Vermont (*W. W. Eggleston*); Billerica, Massachusetts (*C. W. Jenks* and *C. W. Swan*); Dedham, Massachusetts (*Miss Alice Browne*); Providence, Rhode Island (*George Thurber*); New

[10] Mem. Torr. Bot. Cl. **5**: 280.

York (*J. Carey*); Easton, Pennsylvania (*T. C. Porter*); Farmer Mt., Carroll co., Virginia (*John K. Small*).

Habitally *Pycnanthemum verticillatum* should rarely be confused with either *P. muticum* or *P. Torreyi*. *P. muticum* has broader ovate-serrate leaves, the upper distinctly whitened; the bracts of the glomerules are linear or lance-attenuate; and the long stamens are generally much exserted. Though in foliage *P. verticillatum* is approached by *P. muticum*, var. *pilosum* Gray, that form is readily distinguished by its characteristic pubescence and bracts.

From the somewhat similar *P. lanceolatum*, to which Bentham once referred it, *P. verticillatum* is distinguished by its broader leaves, the upper ones pubescent; by its more open inflorescence; by the longer more acuminate and less pubescent bracts of the glomerule; and by the sharper calyx-teeth and smaller corolla.

Some forms of *Pycnanthemum Torreyi* approach *P. verticillatum*, but usually that species may be readily told by its thinner narrowly lanceolate leaves, distinctly tapering at the base, the upper mostly glabrous; and by the narrower bracts of the glomerule. The stamens of the two plants present a striking difference. In *P. Torreyi* they are long-exserted, equaling the upper lip of the corolla; while in *P. verticillatum* they are scarcely 1^{mm} long, and included within the throat of the corolla. It is possible that this is a case of heterostyly, which is rarely observed in *P. lanceolatum*, and that the two plants should be considered extreme forms of one species. From the facts, however, that this floral character, in all the specimens examined, is accompanied by definite characters of pubescence, leaf, and bracts; that *P. verticillatum* extends much further north than the narrow leaved *P. Torreyi*; and that, in one point or another, *P. verticillatum* approaches equally near to *P. lanceolatum*, *P. muticum*, and *P. Torreyi*, it seems more reasonable to treat it as a species. The specific characters in the genus Pycnanthemum are not so fixed as would be convenient for arbitrary diagnoses; but as species go in the group, *P. lanceolatum* and *P. linifolium*, *P. muticum* and *P. incanum*, for example, *P. verticillatum* seems worthy of recognition.—M. L. FERNALD, *Gray Herbarium, Cambridge, Mass.*

THREE NEW CHORIPETALAE FROM NORTH AMERICA AND MEXICO.

Silene rectiramea. — Cespitose perennial, 2 to 3^{dm} high: stems several from a multicipital caudex, covered at the base by the pale scarious persistent scale-like ciliated bases of the earliest leaves, terete or elliptic in section, slightly striate in a dried state, pubescent and more or less viscid especially above, sometimes simple to the inflorescence, sometimes branched from every node; lower internodes relatively short, often curved, the middle and upper elongated, 6 to 8^{cm} long, much exceeding the leaves, remarkably straight; branches solitary or opposite at the nodes, diverging from the stem at a uniform angle of about 45°; their internodes also elongated and very straight: leaves of the stem about five pairs; the lower ones, like the radical, oblanceolate, 2.4 to 4^{cm} long, 4 to 7^{mm} broad, the middle and upper lance-oblong to linear, all acute, 1-nerved, obscurely pinnate-veined, minutely papillose and pulverulent-puberulent under a strong lens: bracts lance-linear, often purplish, 7 to 9^{mm} long; bractlets similar, 2^{mm} long: flowers terminal on the divergent branches of and open flat-topped cyme, or in weaker stems reduced to a terminal and one or two short-pediceled lateral ones; calyx cylindric in anthesis, white and papery but veined with light purple, 9^{mm} long, 10-nerved; the nerves opposite the teeth branching freely, intermediate ones subsimple; teeth orbicular with incurved margins: gynophore in anthesis 1.7, in fruit 2.5^{mm} long: petals 1 to 1.1^{cm} long, glabrous except externally at the very base; claws patulate, subauriculate at the summit, 3-veined; blade short, 2 to 3^{mm} long, bifid a fourth of its length: stamens 10, equal: carpels 3; capsule ovoid, 7^{mm} long, at maturity 1-celled to the very base: seeds reddish-brown, tubercles in few concentric series, those of the dorsal region enlarged and forming a more or less definite crest.

Collected by Professor D. T. MacDougal about the Grand Cañon of the Colorado in Arizona, altitude 2150^{m}, 28 June, 1898, no. 181. Type in herb. Gray.

This species stands near *S. verecunda* Watson, but differs in its elongated very straight branches and delicate papery calyx, which, although narrowly cylindric to obovoid, shows no indication of the tightening or constriction about the carpophore which is to be noticed in *S. verecunda*. Specimens of *S. rectiramea* were distributed in Mr. MacDougal's interesting Arizona sets, but were determined only to the genus. Mr. A. A. Heller, who has had charge of the identification of the sets, has courteously waived in favor of author his right to characterize this species.

Arabis Crandallii. — Cespitose perennial, 3^{dm} high, pale green and hoary puberulent throughout, with minute stellate interplexed hairs: stems numerous (20 or more), slender, terete, from a loosely multicipital caudex: root single, vertical: radical leaves oblanceolate-spatulate, 1.5 to 1.8^{cm} long, 3 to 4^{mm} broad, entire, acutish, cuneate-attenuate at the base, concolorous, minutely stellate-tomentulose on both surfaces, 1-nerved; the cauline (about 8 on each stem) similar but shorter and more oblong, sessile by a subamplexicaul base: pedicels ascending or appressed, 5 to 6^{mm} long, slightly enlarged at the summit: sepals oblong, obtuse, stellate-puberulent, often purplish-tinged, 3^{mm} long: petals obovate, cuneate, white, roseate-tinged, twice as long as the calyx: pods erect, slender, subtorulose in dried specimens, 2.5 to 4^{cm} long, 1^{mm} broad, flattened; seeds (immature) uniseriate in each cell and nearly or quite as broad as the septum.

Collected by Professor C. S. Crandall at Cimmarron, Montrose co., Colorado, attitude 2100^{m}, 18 May, 1898, no. 6. Type in herb. Gray.

In habit this species most nearly approaches the Canadian *A. Macounii* Wats., from which, however, it is clearly distinguished by its fine stellate pubescence, shorter erect pods, and larger leaves.

Mimosa acapulcensis (Subg. *Eumimosa* Ser. *Sensitivæ*). — Shrub 1.5 to 3^{m} high: branches terete, subflexuous, covered with a yellowish-gray striate cortex and armed with scattered, firm, grayish-brown, laterally compressed, slightly recurved spines: leaves unijugate and pinnæ 3-foliolate in the manner of *M. sensitiva* (a diminutive fourth leaflet occasionally present); leaflets glabrous, glaucous, coriaceous, oblong, entire, cartilaginous-margined, acute to rounded at the apex, subcordate and very oblique at the base, oblong, 3 to 10^{cm} long, nearly half as broad; common petioles slender, wiry, 3 to 8^{cm} long; secondary rhachises 2 to 3.5^{cm} long: peduncles slender, ascending, fascicled by 3's, 4's, and 5's along the terminal portion of the branch, and forming a loose, elongated, relatively narrow inflorescence, leafy towards the base: heads globose, 1.5^{cm} in diameter (incl. the long stamens), roseate; flowers perfect and staminate: calyx campanulate, less than 1^{mm} long, cuspidate-denticulate: corolla 2.7^{mm} long, glabrous, 3–4-nerved and 3–4-toothed; teeth ovate-deltoid, a third as long as the tube: pods 2.5 to 4^{cm} long, glabrous and wholly unarmed both as to valves and replum, long-stiped, torose, acuminate at the tip, 3–4-jointed, Indian-brown at maturity; valves falling away in segments.

Common on hills near Acapulco, Mexico, where collected by Dr. Edward Palmer between October 1894 and March 1895, no. 296.

I am indebted to Dr. Rose for calling my attention to this species, Dr. Palmer's specimen having been undistributed in the Gray Herbarium at the time of my recent revision of the genus. Types in herb. Gray and herb. U. S. National Museum. — B. L. ROBINSON.

THE PROBABLE CAUSES OF THE POISONOUS EFFECTS OF THE DARNEL (*LOLIUM TEMULENTUM* L.).

THE presence of a poisonous principle in the darnel has been well known since the earliest investigations of the subject, and recent experiments confirm this fact. According to Hofmeister,[1] the darnel contains two active principles: *temulin*, obtained by this author as chloroplatinate, which acts upon the nervous system; and the other, determined by the oily substances and fatty acids which are contained in the seed in large proportion, which attacks the alimentary canal.

In the course of our researches upon the seed integuments and the pericarp of grasses, we have had occasion to note the practically constant presence in the seeds of the darnel of a fungus to which it seemed reasonable to us to assign the poisonous effects. This fungus, which is always present in the form of mycelial filaments, appears at an early stage in the interior of the ovary. In the first stages of its development it invades the entire nucellus. At the time that the external integument of the ovule disappears, the nucellus itself is almost entirely resorbed, with the exception of two or three of the outermost layers which, obliterated in the maturing of the grain, constitute the hyaline layer. Crowded out by the development of endosperm after fertilization, the mycelium becomes restricted between this hyaline layer and the outermost endosperm. It is in this zone that we have observed it in the mature seed. After the removal of the diverse coatings of the fruit, the hyphæ which constitute this fungus zone appear as septate filaments, generally very long, more or less branched and interwoven with one another. We have found this disposition of the fungus in material from Bolivia, Brazil, Chili, Abyssinia, Persia, Syria, Spain, Portugal, Sweden, Germany, and many localities in France. In forty seeds of most diverse origin the mycelial zone was lacking from but

[1] Archiv. f. exp. Path. u. Pharm. **30**: —. 1892.

three. This observation has been confirmed in other species of Lolium, to wit, *L perenne* L., *L. arvense* With. (var. of *L. temulentum*), *L. linicola* Sond. It is only exceptionally that the first of these contains the parasite. The rest are infected to the same degree as *L. temulentum*. When one observes that the species reported poisonous are the very ones in which we have found the parasite, it seems reasonable to ask whether the temulin of Hofmeister is not a result of the action of the fungus upon the nitrogenous materials in the peripheral region of the seed.

This fungus, of whose nature we are not yet satisfied, may not in any case be identified with the *Endoconidium temulentum* of Prillieux and Delacroix. The latter attacks the seeds of rye which it clearly deforms, the infected grains becoming smaller and lighter than the normal ones. The grains of the darnel show no such deformation. Further, in the rye grains thus attacked, and called "*seigle enivrant*," the protecting layer has generally disappeared, and all the external part of the endosperm has been invaded by the parasite. In the darnel the endosperm suffers no alteration from the action of the fungus, the protecting layer itself remaining perfectly intact. Since our observations were made the same mycelial layer has been noted by Hanausek and Nestler,[2] and before them by Vogl.[3]

Our results are practically alike. However, the other authors have described the fungus for *L. temulentum* alone. — P. GUÉRIN, *Préparateur à l'École Supérieure de Pharmacie de Paris.*

[2] Berichte der deutschen botanische Gesellschaft **8**: —. *pl. 12–17.* November 30. 1898.

[3] Die wichtigsten vegetablischen Nahrungs- und Genussmittel. 1898.

OPEN LETTERS.

IN YOUR June number and in a *Bulletin* of the South Carolina Agricultural Experiment Station, Dr. A. P. Anderson quotes me as identifying the Tilletia found on rice in South Carolina with *Tilletia corona* Scrib. The resemblance is certainly striking, but in writing Dr. Anderson I did not intend to express a final opinion in the matter. I had not at that time seen a description of the Japanese *Tilletia horrida* Tak. It now seems to me that the differences in the manner of affecting the host plant, the spore mass being included by the glumes in *T. horrida* and conspicuously exserted in *T. corona*, should be considered of sufficient weight to separate the species tentatively, until such time as their life histories can be carefully studied. Therefore I should prefer to call the South Carolina specimens *Tilletia horrida* Tak. In my opinion much more confusion is occasioned by the hasty grouping of many forms under one common name than by tentatively recognizing too many forms as independent species.

The name *T. corona* Scrib. seems to be antedated by Arthur's *Ustilago rotundata* (Prel. List Iowa Uredinæ, Nov. 1884), described from Iowa specimens on *Panicum irrigatum*. This species has recently been issued as *no. 543* of *Economic Fungi*, under the name *Tilletia rotundata* (Arth.) Ell. & Ev.—F. S. EARLE, *Auburn, Ala.*

CURRENT LITERATURE.

BOOK REVIEWS.

The Sandusky flora.

A LOCAL flora of much interest has been published by the Ohio Academy of Science as the first of a series of "special papers," the second and third numbers of which are devoted to insects. This initial effort reflects credit upon the Academy, both for the subject-matter and for the manner of its presentation. The volume of 167 pages is thoroughly satisfactory in its typographical and press work, and records a piece of careful local exploration worthy of the excellent setting.

The flora of Sandusky and vicinity as here presented is the result of the labors of Mr. E. L. Moseley[1] during the last seven years, and is based upon the herbarium of the Sandusky high school, while specimens of most of the rarer forms are also deposited in the Gray herbarium of Harvard University and in the herbarium of the Ohio University.

The catalogue proper, which is well indexed, is preceded by thirty-four pages of text, in which a number of interesting matters pertaining to the flora are discussed. It adds not a little interest to learn that the flora of Sandusky is exceptional in the number of its species, having more than is given in those excellent floras by David F. Day for the Buffalo region, by Wm. R. Dudley for the Cayuga region, and by the local Academy of Science for the Rochester region, although these regions border on the great lakes and are severally larger than that included for Sandusky. Even the whole of England does not possess a hundred species of phanerogams more than are found about Sandusky. The islands included in the area, a half dozen having been explored and studied with special care, do not add materially to the richness of the flora, as they have only a few plants not found on the mainland. The surprising abundance of species is ascribed to favorable physiographical conditions in part, but more especially to the climate. The summer is longer at Sandusky than at other points along the shore of Lake Erie, being much over a month longer than at Buffalo, and the spring and summer have a higher average temperature. The reasons for this state of things are discussed and statistical evidence presented.

[1] MOSELEY, E. L.: Sandusky flora, a catalogue of the flowering plants and ferns growing without cultivation in Erie county, Ohio, and the peninsula and islands o Ottawa county. Special paper No. 1. Ohio Academy of Science; Wooster, 1899. 8vo, pp. 167, 1 map.

The nomenclature of the catalogue is that of the *Kew Index*, with some concession to the last edition of Gray's *Manual*. The enumeration begins with *Botrychium ternatum* Swartz and ends with *Xanthium Canadense* Mill. Altogether it is an excellent catalogue, and reflects credit upon the industry and ability of the author, and upon the enterprise of the Academy of Science. —J. C. A.

Anatomy of the dicotyledons.

A SENTENCE of Radlkofer, which the author thinks prophetic — "The next hundred years belong to the anatomical method" — inspired Solereder to bring together the immense mass of material regarding the anatomy of the stem and leaves of the dicotyledons into a handbook for botanical laboratories.[2] To the previously accumulated knowledge of anatomy the author himself has been a notable contributor.

The labor of compilation alone must have been immense. Here are brought together in systematic form the data regarding each of the families of dicotyledons. The author first presents a synoposis of the chief anatomical characters of the family as a whole; then gives an extended account of the leaf structure, followed by a similar presentation regarding the stem. Each section closes with a thorough bibliography.

In an introductory chapter Dr. Solereder explains what is meant by the anatomical method, and discusses the more important anatomical characters and their value in taxonomy. An extensive closing chapter (75 pages) is devoted to a synopsis of the various anatomical features, with reference to their occurrence in certain families, genera, and species — a sort of comparative anatomy.

Though one may doubt whether the anatomical method is destined to play the important rôle in taxonomy which Radlkofer and his pupil Solereder believe, and may easily find matter for criticism in the introduction, as well as flaws in the details regarding structure, the enormous labor which the author has performed in the production of the work disarms criticism and evokes only praise. The book is unquestionably a most useful one for reference, not only to the systematist, but to the histologist and to the physiologist as well. It will prove indispensable in every botanical department where active work is in progress and will doubtless demonstrate its value from day to day.

As a reference book it has one serious defect, the want of a full index. The index only includes the families, and while ordinarily this will give the desired clue, the value of the book would be greatly enhanced by an entry of

[2] SOLEREDER, HANS: Systematische Anatomie der Dicotyledonen. Ein Handbuch für Laboratorien der wissenschaftlichen und angewandten Botanik. 8vo. pp. xii + 984. *figs. 189*. Stuttgart: Ferdinand Enke. 1898–1899. *M* 12.

each genus and species and of each author mentioned. The Bavarian Academy of Sciences, which has assisted in its publication, might well have increased its subvention if necessary to provide such an index.—C. R. B.

Speculative biology.

In 1875 Pflüger propounded a hypothesis regarding the constitution of organized bodies which may be described as the hypothesis of chemical continuity. Impressed with the extensive polymerization among carbon compounds, especially the proteids, he ventured the suggestion that in an organism polymerization may progress indefinitely, so that the whole protoplasm is not an aggregate of similar molecules having definite molecular weight but may form a single giant chemical molecule. This theory has found few adherents. It is accepted *in toto*, however, by Dr. Georg Hörmann, who proceeds in a recent book[3] to show its adequacy to explain certain biological problems, and, therefore, its inherent probability.

He applies it to the transmission of the impulse in nerve and the phenomena of nerve section; to the contraction of muscle and the discharge from the electric organs of fishes; and discusses the structure of the cell and the rotation of the protoplasm "from the standpoint of the *principle* of chemical continuity." (Hypothesis — principle: are they synonymous?)

Of course the book is pure speculation, and must not be taken as anything else, though we fear the author does not always remember the sandy foundation on which he is building. The various ingenious diagrams, representing atoms of divers interesting forms and positions lend an air of verisimilitude which might deceive the very elect.

Theory we recognize as necessary; speculation is indispensable in the formation of multiple working hypotheses by the investigator; but it may be seriously doubted whether the publication of a speculation is ever worth while. Until we have more intimate knowledge of the chemistry of proteids, speculation of the kind here set forth must be regarded as little more than vanity and vexation of spirit.—C. R. B.

NOTES FOR STUDENTS.

Continuing his observations on the agencies by which insects are attracted to flowers, Professor J. Plateau now gives a large number made on *Salvia horminum* and *Hydrangea opuloides*,[4] confirming his previous statement that they are chiefly attracted by the sense of sight. Neither the col-

[3] Hörmann, Georg: Die Kontinuität der Atomverkettung ein Strukturprinzip der lebenden Substanz. 8vo. pp. iv + 118. *figs.* 32. Jena: Gustav Fischer. 1899. *M* 3.

[4] Mém. Soc. Zool. de France **11**: 339–375. *fig.* 4. 1898.

ored bracts in the former nor the conspicuous sterile flowers in the latter plant can be regarded as "vexillary." In both cases the pollinating insects make their way at once to the flowers which contain the honey without being visibly guided by the showy organs in either case; while if these are removed it does not appear to make any material difference in the number of insects which visit the inflorescence.— Jour. Roy. Mic. Soc. 1899. 298.

IN A NOTABLE paper on cellulose enzymes, Professor F. C. Newcombe clearly demonstrates the existence of cytohydrolytic enzymes distinct from diastase, especially in the seedlings of white lupine and date palm.[5] These enzymes, which in some plants are doubtless mixed with diastase, "act on starch so feebly and on reserve cellulose so energetically that they are to be regarded as cytase as distinguished from diastase." In all cases the cell walls first become hyaline, then more and more transparent, finally seeming to melt away in solution. Besides the clear proof of the existence of the long suspected cytase, the paper adds much to our knowledge of the distribution of cellulose enzymes.— C. R. B.

DR. A. M. BOUBIER in a brief paper on the pyrenoid[6] states that his observations "prove the existence in pyrenoids of an external plasmic membrane, perfectly differentiated and independent of the chromatophore, at least at its mature stage of development. This membrane encloses a leucoplast, which accumulates starch, with a crystalloid at the center.— C. R. B.

DR. G. HOCHREUTINER has determined experimentally that seeds of a number of aquatic plants may pass uninjured through the alimentary canal of herbivorous fish, and the latter may therefore aid in the dissemination of aquatic plants.[7]— C. R. B.

THE CHIEF VALUE of Kolkwitz's recent paper on the influence of light on the respiration of fungi[8] is due to the refinement of technique employed and the degree of accuracy attained. In these respects it surpasses previous work and eliminates many sources of error that have been neglected. This is the first extensive accurate study of the effect of light upon the respiratory activities of plant protoplasm and of animal protoplasm as well. Animals are prone to move and then by their varying activities to render invalid any conclusions as to the effect of light alone. Severed parts of plants are unsuitable, as diffusion at the cut end is abnormal and quantitatively altered. Fungi having false parenchyma are unsuitable since the intercellular spaces

[5] Annals of Botany **13**: 49-81. 1899.

[6] Bulletin de l'Herbier Bossier **7**: 451-458. 1896.

[7] Bulletin de l'Herbier Boissier **7**: —. 1899.

[8] KOLKWITZ, R: Ueber den Einfluss des Lichtes auf die Athmung der niederen Pilze: Jahr. f. wiss. bot. **33**: 129-165.

may suffer change from light. Therefore the author selects such fungi as produce a loosely woven mycelium that spreads itself out openly to the light (Aspergillus, Penicillium, bacteria), and measures such activities as are solely dependent on light. As sources of error he recognized chiefly the evolution of CO_2 through decomposition of oxalic acid or of dead parts, and errors introduced by variation in temperature. The classical method of Pettenkofer (1862) is adopted and the amount of evolved CO_2 is determined by titration with oxalic acid. To secure greater accuracy the gas was forced, not drawn, through the apparatus at the rate of three, four, or five liters per hour as desired. The process and apparatus are described in great detail. Suffice it to say here that exceeding care was given to every feature. The culture vessel, of special design, presented a great surface to the light while of but small capacity. In order to reduce the absorption by glass the walls of the vessel were very thin. The feature wherein this study chiefly surpasses previous work is in the regulation of the temperature of the culture during experimentation. This was accomplished by immersing the culture vessel in a tank containing six liters of water and keeping it at a constant temperature by electricity, automatically regulated by a very ingenious contrivance. Lest the thin layer of water covering the culture flask should vary in temperature all of the water was continually agitated by a paddle operated by a turbine. The inflowing air was warmed to the temperature of the water. In this way the variation was from one tenth to one thirtieth of a degree C. The electric light was constant in quantity and quality, thus avoiding the variations inevitable in the use of sunlight. Estimations of CO_2 were made every two minutes. The author announces as a result of his labors that light, under the conditions employed, increases respiration about 10 per cent. The effect is observable in young or old cultures, richly or poorly nourished fungi, and in acid or alkaline media. The influence of light during long periods when secondary processes arise was not investigated. An excellent bibliography is given.—F. L. STEVENS.

ITEMS OF TAXONOMIC interest are as follows: In continuing his flora of the West Indies (*Symbolæ Antillanæ*), URBAN, in the second part, presents the Araliaceæ, represented by four genera and sixteen species; and in another paper describes about eighty miscellaneous new species, chiefly from Puerto Rico, among which we notice two new genera, *Notodon* (Leguminosæ, near Sabinea) and *Hybosperma* (Rhamnaceæ). LINDAU presents the Polygonaceæ, represented by ten genera and sixty-six species, the great genus Coccoloba containing about fifty of them. SCHLECHTER presents the Asclepiadaceæ, represented by twenty-one genera and eighty-eight species, the largest genus being Metastelma, with thirty-four species, eighteen of which are new. Two new genera are established, *Tainionema* and *Decastelma*.—K. MIYAKE (*Bot. Mag. Tokyo* **13**: 1–4. *pl. 3*. 1899) has described a new genus of Hepaticæ,

very closely resembling Pellia. It is said to have a spermatozoid much larger than that of Pellia, which has heretofore been credited with the largest spermatozoids among the Hepaticæ. The new genus is known as *Makinoa*, in honor of Makino the discover. Specimens without sporogonia had already been described by Stephani as *Pellia crispata*, so that the name stands as *M. crispata* (St.) Miyaki.— In *Proc. Amer Acad.* (**34**: 507–534. 1899) ROBINSON and GREENMAN publish revisions of Montanoa, Perymenium, and Zaluzania, long a puzzling series of Mexican and tropical American composites. *Montanoa* is recognized as containing thirty-two species, nine of which are new; *Perymenium* has twenty-six species, ten of which are new; and *Zaluzania* has twelve species, two of which are new. The same authors (*ibid.* 534–566) have published a synopsis of the genus *Verbesina*, which has not been treated as a whole since 1836 (DC. Prodr.), at which time thirty-three species were recognized, all but two being American. Now the genus is conceded to be exclusively American, and contains 109 species, more than 70 per cent. of which are local. The greatest display of species is in the uplands of central and southern Mexico, where 40 per cent. of the species are endemic. In the synopsis twenty-five new species are described.— GREENMAN (*ibid.* 566–578) has published some new and critical Mexican species, the new species numbering twenty.— ELIAS NELSON has published a revision of the phloxes of western North America. He recognizes thirty-eight species, nineteen of which are new. The paper is a master's thesis in the University of Wyoming, and is published in the ninth report of the Wyoming Agricultural College, Laramie, Wyoming.— E. P. BICKNELL, in continuing his studies of Sisyrinchium (*Bull. Torr. Bot. Club* **26**: 297–300. 1899), has described four new species from Michigan.— A. A. HELLER (*ibid.* 312–315) has described additional new species from western North America.— AVEN NELSON (Erythea **7**. 57–64. 1899) has discussed the western species of *Aragallus* (*Oxytropis*), describing eight new species; and has also described (*ibid.* 65–70) five new forms of Oreocarya, two of Cryptanthe, and one of Allocarya.—JARED G. SMITH (Bull. 18. Div. Agrost. U. S. Depart. Agric.) has published a synopsis of the genus *Sitanion*, recognizing twenty-three species, twenty of which are new.—J. M. C.

GUIGNARD has recently studied the reduction of chromatin in *Naias major*[9]. This plant has proved exceptionally favorable for such a study, since the number of chromosomes, twelve in the sporophyte and six in the gametophyte, is the smallest yet reported for any flowering plant.

First division.—In the prophase the spirem splits longitudinally and then segments into six primary chromosomes each of which consists of two pieces. During the succeeding contraction and growth, each of these pieces shows a

[9] Le développement du pollen et la réduction dans le *Naias major*. Arch. d'anat. microscopique **2**: 455–509. 1899.

double row of chromatin granules, a preparation for a second splitting, so that the primary chromosomes are to be regarded as quadruple. As the primary chromosome separates into its two parts (secondary chromosomes), the splitting already inaugurated by the fission of the granules begins to take place, but is not entirely completed, since the two chromosomes remain united at their extremities, thus forming a V with its apex attached to the contractile threads of the spindle. Each daughter nucleus receives six double (secondary) chromosomes.

Second division.—In the second division, six V-shaped chromosomes appear. At the point of the V there is an interruption in the linin support and everything favors the conclusion that these are the secondary chromosomes of the first division which have not lost their individuality. No longitudinal division takes place at this time, there being merely a distribution of the two parts of the V-shaped double chromosome. Thus the two divisions merely distribute the four parts of the primary quadruple chromosome (tetrad), which were already defined in the prophase of the first division. It is evident that there can be no qualitative reduction.

Miss Sargant [10] both figured and described a second fission of the chromatin granules in *Lilium Martagon*, and called attention to the quadruple nature of the primary chromosome. Guignard believes that his results agree with Farmer's account of Pallavicinia, Brauer's of Ascaris, Meves' of Salamandra, and also with Belajeff's description of Iris, although that writer has given a different explanation of the origin of the chromosomes in the pollen mother cell. An excellent review of the chromosome problem, illustrated by very clear diagrams, is by no means the least important part of the work.

The brilliant results of Nawaschin and Guignard on the fertilization of Lilium have been confirmed by Miss Sargant,[11] who finds that during fertilization the male nucleus is applied to the female nucleus, while the second male nucleus is applied to both the polar nuclei. In one case, in which the polar nuclei were not yet in contact, the much elongated "antherozoid" united them like a bridge. In several preparations it was noted that the pollen tube, after fertilization had taken place, contained two small nuclei. Since both generative nuclei are already accounted for, it is suggested that these are probably due to the division of the tube nucleus.—CHAS. J. CHAMBERLAIN.

[10] Ann. Bot. **11**: 187–224. 1897.

[11] On the presence of two vermiform nuclei in the fertilized embryo-sac of *Lilium Martagon*. Proc. Royal Soc. **65**: 163–165. 1899.

NEWS.

A BIOGRAPHY and bibliography of the late Professor William Nylander appears in the June number of the *Revue général de Botanique* (11 : 218–237).

IN AN APPENDIX to the *Bulletin* of miscellaneous information issued by the botanical department of Trinidad, the superintendent, Mr. J. H. Hart, is giving a description of the West Indian and Trinidad ferns.

MESSRS. CARLTON R. BALL, of Iowa Agriculture College, Elmer D. Merrell, of the University of Maine, and P. Beveridge Kennedy, of Cornell University, have been appointed assistants in the Division of Agrostology, U. S. Department of Agriculture.

DR. FRANCIS RAMALEY, of the University of Minnesota, has been appointed professor of biology in the University of Colorado, at Boulder, in succession to Professor John Gardiner, who has retired on account of continued ill-health, having held the chair since 1889.

THE Académie Internationale de Géographie Botanique has conferred its international scientific medal upon Professor John M. Coulter. This medal is awarded by the Academy "in recognition of services rendered to science by those who cultivate and advance its various branches."

A WORK by Dr. C. B. Davenport, of Harvard University, intended especially for botanists and other naturalists who are interested in the quantitative study of species and of organic variation, is announced by John Wiley & Sons, New York City. It is entitled "Statistical methods with specia reference to biological variation."

PROFESSOR DR. EDUARD JANCZEWSKI, Cracow, Austria, is engaged in a study of the geographical distribution and the forms of *Ribes rubrum*. He desires to examine the forms growing in the United States, and particularly the var. *subglandulosum*. Anyone who can furnish a range of herbarium specimens and viable seeds of these forms will confer a favor by communicating with Professor Janczewski.

A "MANUAL of the flowering plants of Iowa," by T. J. Fitzpatrick, of Lamoni, Ia., is announced. The publication of this work is a private enterprise, and intending purchasers should address the author. Part I, containing the Polypetalæ, is not a mere list, but each species is briefly described, together with notes on habitat and distribution, with suitable analytic keys.

A SERIOUS DISEASE of the peach appeared in Michigan and other states several years ago and seems to be spreading. It is known to growers as "little peach" for the reason that the fruit of the affected trees seldom attains more than one fourth normal size. The Division of Vegetable Physiology and Pathology of Washington is making a study of the disease this year, the work being placed in charge of Mr. Merton B. Waite, who is now in the field.

MR. HERMANN VON SCHRENK has been made a special agent in the Division of Vegetable Physiology and Pathology of Washington and authorized by the Secretary of Agriculture to make such studies of the diseases of forest trees and timber as may be directed by the chief of the division to which he has been appointed. This important line of work has been much neglected in this country, and it is hoped that with the start made it will soon have the attention it deserves.

MR. W. A. ORTON, who has for several years been associated with Professor L. R. Jones of the University of Vermont, has been made an assistant in the Division of Vegetable Physiology and Pathology, U. S. Department of Agriculture. Mr. Orton will spend the greater part of the summer in South Carolina and other southern states, investigating the diseases of cotton. He is now at James island, S. C., making some preliminary studies of a disease of sea island cotton which has prevailed in that section for several years.

MRS. ARVILLA BACON ELLIS, wife of Mr. J. B. Ellis, of Newfield, N. J., died at her home on July 18, after a long illness, at the age of sixty-eight. While Mrs. Ellis was not known as a botanist, it was her cooperation which made possible the issue by her husband of the two great sets of fungi exsiccati, the *North American Fungi* and *Fungi Columbiani*. The 3000 portfolios in which the sets were mounted were made by her own hands; and she arranged and folded the pockets for about three fourths of the 200,000 specimens sent out in those books. Her labor of love certainly deserves public recognition.

AN INTERNATIONAL conference on hybridization was held in Chiswick Garden, London, July 11 and 12, under the auspices of the Royal Horticultural Society. An exhibition of hybrids was given on the first day in the large vinery in the Gardens. The conference was attended by many distinguished botanists and horticulturists. Mr. H. J. Webber was the delegate from this country. With the aid of lantern illustrations he described the hybridization work which he and Mr. Swingle are prosecuting under the charge of the Department of Agriculture. Professor L. H. Bailey sent a paper, but was unable to be present in person.

THE YALE CORPORATION, at a recent meeting, voted that the chair of botany held by the late Professor Daniel C. Eaton until his death should

hereafter be known as the Eaton Professorship of Botany. It may be recalled that this professorship was founded and endowed by a relative and by friends of Professor Eaton in the year 1864, although it has never been distinguished by a name. The extensive and valuable botanical library and herbarium which Eaton accumulated have been donated to the university by his family, and Mrs. Eaton has placed in the botanical laboratories a bronze tablet to her husband's memory, and, in addition, has founded a graduate scholarship in botany.—*Erythea*, for August.

PROFESSOR BESSEY has been called upon to serve as the Acting Chancellor of the University of Nebraska, and as the duties of the chancellorship will make it necessary for him to be relieved of much of his botanical work in the laboratories and lecture room, the regents have elected Dr. August L. Rimbach (Ph.D., Jena 1887) *ad interim* Instructor in vegetable physiology and pathology. Dr. Rimbach is known as the author of many botanical papers in the *Berichte der deutschen botanischen Gesellschaft* and other German botanical journals. He was a pupil of Stahl, Detmer, Schwendener, and Sachs, and was Professor of Botany in the University of Cuenca, Ecuador, from 1889 to 1894. He has traveled extensively in South America, engaged in the study of flora of tropical and alpine regions.

MR. J. N. ROSE has just returned from a three months' trip through Mexico, bringing about 900 species of dried plants, many living plants, especially agaves, and plant photographs. His collection is not so large nor so rich in new species as the one of 1897, but it will doubtless help to clear up many puzzles which have long worried botanists dealing with Mexican plants. Besides rediscovering *Echinocactus Parryi*, he collected several other species lost or hitherto unknown to American herbaria. About 200 species were collected at type localities. Among the places visited were Chihuahua, Aguas Calientes, Jalapa, City of Mexico, Oaxaca, Cuernavaca, Pachuca, Real del Monte, Guadalajara, Tequila, San Luis Potosi, and Guanajuato; all places from which many types have been taken. Mr. Rose also made a very thorough study of the species of agave, especially those used in the manufacture of pulque and mescal. The visit to Tequila was chiefly to learn what plant furnishes the "tequila" which is the great mescal drink of Mexico.

Mr. Rose was accompanied by Dr. Walter Hough of the National Museum.